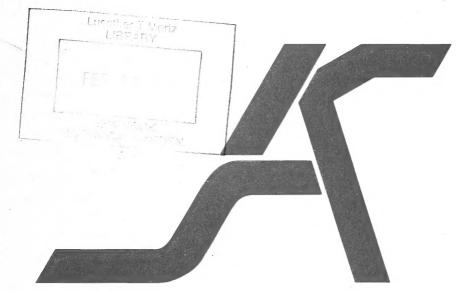
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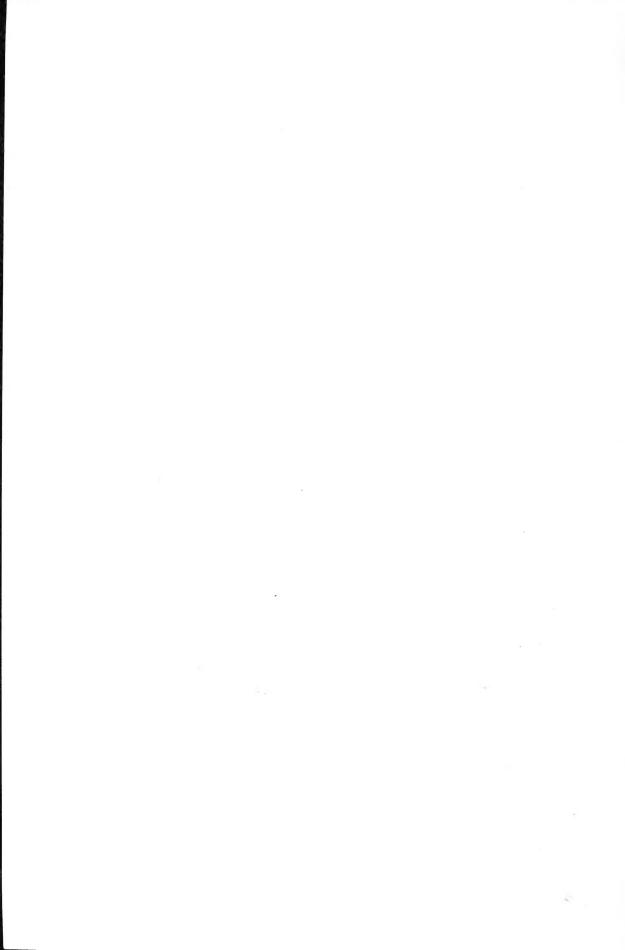
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## Conservation Status and Nesting Biology of the Endangered Duskytail Darter, *Etheostoma percnurum*, in the Big South Fork of the Cumberland River, Kentucky

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#### ABSTRACT

In September 1995 and May and June 1998 and 1999 we conducted an intensive survey of a middle reach of Big South Fork of the Cumberland River in Kentucky with the goal of finding the duskytail darter, Etheostoma percnurum, a federally endangered species. Seventy-one specimens were observed in a 19-stream km reach from the mouth of Station Camp Creek, Scott County, Tennessee, to the mouth of Bear Creek, McCreary County, Kentucky. Using underwater observation and a kick-seining technique around slabrocks, we concur with others that the primary habitat of E. percnurum includes clear, silt-free pools immediately above riffles where it seeks cover under cobbles and slabrocks. Most Kentucky specimens (31 of 35) and all nests were found in a 3-km reach from just upstream of the mouth of Troublesome Creek to the mouth of Oil Well Branch. On 26 May 1998 five nests were found at two sites. All nests were located immediately above riffles in silt-free glides with slabrock and cobble substrates and were guarded by males. Eggs, deposited in a monolayer on the underside of slabrocks, numbered from 79-103 eggs per nest. Slabrocks with eggs had mean dimensions of  $24 \times 19 \times 4.1$  cm and were located in shallow water (51-70 cm) in areas of low flow (5-14 cm/s). Because of the rarity of this darter within its restricted range in Kentucky, we recommend that it be added to Kentucky's list of protected species as endangered. Morphological comparison of E. percnurum from across its range revealed that specimens from Big South Fork have more lateral-line scales, are larger, and are shaped differently than specimens from other populations. The morphological and biological comparisons, along with zoogeographic evidence, indicate that the Big South Fork population of E. percnurum is an independent evolutionary unit.

#### INTRODUCTION

Etheostoma percnurum (Perciformes: Percidae), the duskytail darter, is one of 18 species in the darter subgenus Catonotus (Braasch and Mayden 1985; Page et al. 1992), a group characterized by a derived spawning habit of clustering eggs in a monolayer on the underside of slabrocks (Braasch and Mayden

1985; Page 1985). It is a member of one of three recognized complexes in the subgenus, the *flabellare* complex, which includes the stripetail darter, *Etheostoma kennicotti*, and the fantail darter, *Etheostoma flabellare*, in addition to *E. percnurum*. The duskytail darter, long known only by its common name, was formally described and distinguished from its

closest relative, the wide-ranging E. flabellare, by R. E. Jenkins in 1994 (Jenkins and Burkhead 1994:877-881). At that time, E. percnurum was known from six relict populations in drainages of the Cumberland and Tennessee rivers: one in Virginia, Copper Creek; and five in Tennessee, Citico Creek, Abrams Creek, Little River, South Fork Holston River, and Big South Fork of the Cumberland River. Populations in South Fork Holston River and Abrams Creek are believed extirpated (Etnier and Starnes 1993; Jenkins and Burkhead 1994). The only known site of occurrence in the Cumberland River drainage is Big South Fork. Because of this relict distribution, the presumed extirpation of two populations, and threats to water quality in streams it is known to inhabit, E. percnurum is listed as Federally Endangered (Biggins 1993). Biggins and Shute (1994), Burkhead and Jenkins (1991), Etnier and Starnes (1993), Jenkins (in Jenkins and Burkhead 1994), Layman (1984, 1991), and Simon and Layman (1995) summarized aspects of life history, development, distribution, and abundance based largely on populations in either Copper Creek or Little River.

Despite numerous and intensive fish collections made over the past 40 years, E. percnurum has been reported from only one locality on Big South Fork, the mouth of Station Camp Creek, Scott County, Tennessee. In 1995, we were contracted by the Kentucky Department of Fish and Wildlife Resources (KDFWR) to conduct a status survey of E. percnurum in the Kentucky reach of Big South Fork. In this paper we document the status of this species in Kentucky by presenting distributional, abundance, reproduction, and recruitment data. We describe the habitat and nesting biology for the species in Kentucky. Finally, we identify unique morphological, behavioral, and ecological features that indicate the Big South Fork population may be an independent evolutionary unit.

#### **METHODS**

Status Survey

From 7–9 Sep 1995 we conducted a nearly comprehensive survey of a middle reach of Big South Fork Cumberland River, Kentucky and Tennessee. We surveyed 14 sites judged to have suitable habitat for E. percnurum in a

19-km reach from the mouth of Station Camp Creek, Scott County, Tennessee, to the mouth of Bear Creek, McCreary County, Kentucky. Four of these sites were resampled and two new sites in Big South Fork were surveyed in spring 1998 and 1999 during trips designed to gather information on the nesting biology of E. percnurum. Near the Tennessee-Kentucky border, Big South Fork is a medium-sized river, 30-50 m wide, that flows through a deep (200-300 m) gorge of the Cumberland Plateau. Pools are long and deep, with housesized boulders and bedrock substrates; riffles are fast, well defined, and flow over a substrate of cobbles, boulders, and some pea gravel and coarse sand. The mainstem has a completely forested riparian zone and is protected as a National River and Recreation Area under management of the National Park Service. Because of limited access in this area, nearly all

sites required travelling via canoe.

Underwater visual sampling with snorkeling gear was used at all sites for locating individuals of E. percnurum. As many as 10 people were involved in underwater sampling at one time, thus increasing the efficiency of the search. In suitable habitat, snorkelers fanned out and turned over slabrocks in pools above and below riffles, macrohabitats known to harbor the species (Jenkins and Burkhead 1994; Layman 1991). This method was supplemented at selected sites by kick-seining (Jenkins and Burkhead 1994) isolated rocks serving as potential cover for E. percnurum. About 20-60 minutes of snorkel and/or kick-seining time were spent at each site. Standard physical habitat features (width, depth, area sampled) were recorded at each site. The size of large specimens observed was measured or judged to the nearest millimeter with a plastic ruler. Young-of-the-year (YOY) generally were noted but not measured. Initially, identifications of E. percnurum observed by snorkeling were confirmed by capturing individuals with a dipnet. We quickly discovered that even YOY were easily identifiable while we were snorkeling because of the distinctive appearance of E. percnurum, the only member of Catonotus present in Big South Fork mainstem.

Nesting Biology

Our initial status survey aided in the identification of potential nesting sites for E. perc-

nurum. Six sites were surveyed for nests in a 27-km reach between Station Camp Creek and Blue Heron, McCreary County, Kentucky, on 25-26 May 1998, 24 Jun 1998, and 18 Jun 1999. Underwater visual observation with snorkeling gear was used to locate nesting adults. Snorkelers concentrated on appropriate habitat above riffles, turning over rocks suitable for use as nesting substrate. About 60-230 minutes of snorkeling time were spent at each site, and up to six people were involved in underwater visual observation. All E. percnurum adults observed were captured with dipnets, measured, photographed, and released. The number of eggs in a nest was counted, and the diameter of eggs, nest rocks, and other physical parameters of nest sites were measured with a small plastic ruler or meter tape. Current velocity was measured over the nest with a Swoffer model 2100 flowmeter at 0.6 of the depth above a nest site.

#### **Systematics**

To better understand the evolutionary units under protection, we examined and compared specimens from the drainages of the Cumberland and Tennessee rivers. Seventeen meristic and 27 morphometric variables were taken from 65 and 39 specimens, respectively, of E. percnurum. Measurements and counts of meristic features follow the methods of Hubbs and Lagler (1974) except that scales above the lateral-line were counted diagonally from the origin of the second dorsal fin. Vertebrae were visualized by the aid of soft x-rays (3A, 30 my, 15 seconds) and were counted using the methods of Jenkins and Lachner (1971). Cephalic lateral pore counts followed the methods of Page (1983).

Truss-geometric protocol (Humphries et al. 1981; Strauss and Bookstein 1982) was used in part to archive body form and included 17 measurements distributed among three sagittal truss cells with appended anterior and posterior triangles. Ten additional measurements were included in the morphometric analysis. Multivariate analysis of the morphometric data was accomplished using sheared principal component analysis (PCA) (Bookstein et al. 1985; Humphries et al. 1981) to eliminate overall size effects. Principal components were factored from a covariance matrix of log-transformed morphometric variables following the

recommendations of Bookstein et al. (1985). Multivariate analyses were conducted with programs available in SAS 6.01 (SAS Institute, Inc. 1985) and as modified by D. L. Swofford.

Preliminary morphometric analysis revealed strong sexual dimorphism and seasonal variation associated with reproduction in the Copper Creek specimens (the only ones collected in the spring). To reduce confounding variation associated with reproduction, we removed the Copper Creek material from the analysis, and only compared material from collections made outside of the breeding season (August–February).

#### RESULTS

Status Survey

We observed 60 individuals of E. percnurum in September 1995 and 11 individuals in May 1998 in the 19 stream km reach of the Big South Fork between the mouth of Station Camp Creek, Scott County, Tennessee, and the mouth of Bear Creek, McCreary County, Kentucky (Figure 1). Environmental conditions were ideal because the river was at base flow, water clarity was excellent (at least 2 m), direct sunlight was present, and water temperature was warm, averaging 22.2°C in September 1995 and 22.5°C in May 1998. All sites sampled in Tennessee produced 1-10 E. percnurum, but only 6 of 10 sites sampled in Kentucky produced individuals, ranging from 1-11 including both adults and YOY (Table 1). Most (31 of 35) individuals observed in Kentucky were in a 3-km reach from just above the mouth of Troublesome Creek to the mouth of Oil Well Branch.

Etheostoma percnurum, not common at any site, was probably the least common darter species observed. Considering that we adequately sampled only 25–50% of the suitable habitat at any one site, the species is presumably more abundant than our results (Table 1) might otherwise suggest. Our professional judgment is that at least 5–10 times the numbers we observed almost certainly inhabit a given site. This conservative estimate would yield a total population of 300–600 individuals of *E. percnurum* in a 19-km stretch of Big South Fork.

Underwater observation proved to be a productive method of finding and observing *E*.

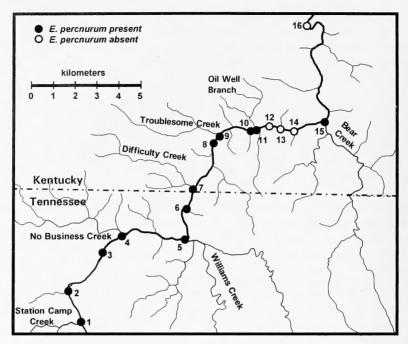


Figure 1. Sampling sites for *Etheostoma percnurum* along Big South Fork of the Cumberland River, Scott County, Tennessee, and McCreary County, Kentucky.

percnurum in most habitats. For comparison, we kick-seined isolated rocks judged to potentially harbor *E. percnurum* at three sites in Kentucky. Initially, our success rate was high, as 3 of 6 rocks sampled yielded *E. percnurum* (site 8). Subsequently, at sites 9 and 11 only 2 of 14 rocks and 1 of 14 rocks sampled, respectively, yielded *E. percnurum*. At these three sites our yield per unit effort for kick-seining (1 fish per 23.3 person-minutes) was considerably higher than for underwater observation (1 fish per 80.8 person-minutes). However, the stacked slabrocks and boulders of most areas precluded sampling by the kick-seining method.

#### Habitat

All E. percnurum were observed in silt-free pools or raceways with low, but evident flow, immediately above riffles where cobbles, boulders, and slabrocks were available. These pools averaged about  $25 \times 40$  m in area and about 54 cm deep, although specimens were observed as deep as 1.5 m. All individuals were under cover of cobbles, boulders, or slabrocks. Cover rocks ranged from fist-sized

cobbles to 76 × 76 cm slabrocks and boulders, with an average thickness of 5 cm. Eleven other darter species were found in association with *E. percnurum*, including *Etheostoma baileyi*, emerald darter; *Etheostoma camurum*, bluebreast darter; *Etheostoma caeruleum*, rainbow darter; *Etheostoma cinereum*, ashy darter; *Etheostoma sanguifluum*, bloodfin darter; *Etheostoma stigmaeum*, speckled darter; *Etheostoma tippecanoe*, tippecanoe darter; *Etheostoma zonale*, banded darter; *Percina caprodes*, logperch; and *Percina copelandi*, channel darter.

#### Nesting Biology

Five nests were located in 44.7 personhours of snorkeling, for a rate of one nest per 8.9 hours of snorkeling. Three nests were found at site 8, just above the mouth of Troublesome Creek, and two nests were found at site 10, mouth of Annie Branch, all on 26 May 1998 (Table 2). All nests were in pools and raceways, 5–50 m above riffles. Nests were in water 51–70 cm (mean = 62 cm) deep with current velocity at 5–14 cm/s (mean = 10 cm/

Table 1. Geographic location, date, number of individuals observed while snorkeling, unit of effort (snorkelers  $\times$  minutes), and approximate size of specimens of *Etheostoma percnurum*, Big South Fork Cumberland River, Kentucky and Tennessee. Geographic location numbers correspond to those in Figure 1. Sexable adults are divided into males (M) and females (F).

	Geographic location	Date	Number of individuals (sex)	Unit of effort	Size of specimens (mm, TL)
1.	Mouth of Station Camp	7 Sep 1995	6	9 × 60	35-50 + YOY
	Creek, Scott Co., TN	25 May 1998	2F	$8 \times 30$	50
2.	Mouth of Parched Corn Creek, Scott Co., TN	7 Sep 1995	6	$9 \times 60$	40–50
3.	Halfway between Cold Spring and Big Branch, Scott Co., TN	7 Sep 1995	10	9 × 60	30–50 + YOY
4.	Big Island, Scott Co., TN	7 Sep 1995	10	$9 \times 90$	35-50 + YOY
5.	Just above mouth of Williams Creek, Scott Co., TN	8 Sep 1995	1	$9 \times 60$	30
6.	Near mouth of Hurricane Creek, Scott Co., TN	8 Sep 1995	1	$9 \times 60$	35
7.	Mouth of Difficulty Creek,	8 Sep 1995	1	$9 \times 60$	40
	McCreary Co., KY	25 May 1998	2	$6 \times 60$	35
8.	1 km above mouth of Trou- blesome Creek, McCreary	8 Sep 1995	11	$9 \times 110$	40–60 YOY
	Co., KY	25–26 May 1998	3M, 1F	$6 \times 180$	50-65
9.	Mouth of Troublesome Creek, McCreary Co., KY	9 Sep 1995	7	$10 \times 75$	30–55 + YOY
10.	Mouth of Annie Branch, McCreary Co., KY	26 May 1998	2M, 1F	$6 \times 35$	54–67
11.	Mouth of Oil Well Branch, McCreary Co., KY	9 Sep 1995	6	$10 \times 20$	40–60 + YOY
12.	Huling Ford, McCreary Co., KY	9 Sep 1995	. 0	$10 \times 20$	
13.	Mouth of second unnamed tributary below Huling Ford, McCreary Co., KY	9 Sep 1995	0	$10 \times 30$	
14.	Mouth of tributary near Slav-	9 Sep 1995	0	$10 \times 30$	
	en's Branch Trail, Mc- Creary Co., KY	26 May 1998	0	$2 \times 20$	_
15.	Mouth of Bear Creek, Mc- Creary Co., KY	9 Sep 1995	1	$10 \times 120$	50 mm
16.	Blue Heron, McCreary Co.,	24 Jun 1998	0	$2 \times 90$	_
	KY	18 Jun 1999	0	$2 \times 150$	

Table 2. Summaries of physical habitat features and nest characteristics of five nests of *Etheostoma percnurum* at sites 8 and 10 (see Figure 1 and Table 1) in Big South Fork Cumberland River, Kentucky, 26 May 1998.

Nest parameter	Mean	Range
Length of nest rock	186 mm	150–300 mm
Width of nest rock	240 mm	180–400 mm
Thickness of nest rock	41 mm	37–50 mm
Depth of nest	62 mm	51–70 mm
Height of nest rock cavity	21 mm	15–25 mm
Size of guardian male	57 mm	55–58 mm
Water temperature	22.5 C	22.5 C
Length and width of egg mass	$40 \times 50 \text{ mm}$	$30 \times 40$ mm– $50 \times 75$ mm
Diameter of eggs	2–3 mm	2–3 mm
Number of eggs in nest	101	79–132
Current velocity	9.6 cm/s	5.0-14  cm/s

Table 3. Frequency distribution of lateral-line scales in four populations of *Etheostoma percnurum*, Virginia and Tennessee.

Population	39	40	41	42	43	44	45	46	47	48	49	50	n	Mean	SD -
Copper Creek, VA	2	1	5	3	5	3	1						20	42.05	1.67
Big South Fork, TN						2	1	1	2	1		1	8	46.38	2.07
Little Rock, TN		3	1	5	6	8	4	2	_	1	1		31	43.55	2.08
Citico Creek, TN			1	1	_	1	1	2					6	44.00	2.10

s). All nest rocks were slab-sided and ranged from  $15 \times 18$  cm to  $30 \times 40$  cm and averaged 4.1 cm in thickness. A cavity 15–25 mm deep was between the substrate (sand, coal, and detritus) and the bottom of the nest rock. Nests were located in a relatively small area at each site; nests ranged from 2.4–7.0 m apart. Water temperature was  $22.5^{\circ}$ C at both sites.

Eggs were adhesive, round, 2–3 mm in diameter, and amber. They were deposited in a monolayer on the underside of the nest rocks except one nest had one egg laid on top of another egg. Four of the five clutches contained "eyed" embryos rapidly moving inside their chorions. The number of eggs in the nest clutches (or the complement of eggs) ranged from 79–132 (mean = 101). Clutches were oblong to round, ranged in size from  $30 \times 40$  mm to  $55 \times 75$  mm, and generally placed near the center of the underside of the nest rock.

Each of the five nests was guarded by a single male with nuptial colors and morphology similar to other members of Catonotus. The knobs on the tips of the first dorsal fin were bright gold to amber, and the edges of the pectoral, soft dorsal, and caudal fins were distinctly peppered with black margins. All males had strong vertical bar development on their sides and blackened heads. The bases of their caudal and pectoral fins were light amber to salmon; pelvic fins were an iridescent white. Standard length (SL) for the five guarding males ranged from 55-58 mm (64-67 mm total length). The four adult females (50–54 mm SL) were found under rocks well away from nests. None of the females were swollen with mature ova and apparently already had spawned.

#### Parasites

In May 1998 we observed black-spot disease in five of six specimens examined closely. The number of black spots ranged from 1–4, except that one female had about 25 spots.

This female was covered in fungus and appeared to be near death. Black-spot disease was present in only three of the eight specimens vouchered in September 1995; the number of black spots ranged from 1–5.

#### Systematics

The Big South Fork population had higher mean lateral-line scales than other populations (Table 3). Specimens from Citico Creek were distinctive in having fewer principal caudal rays, scales above the lateral line, scales below the lateral line, scales around the caudal peduncle, and lateral-line scales and more pored lateral-line scales (Table 4). Other meristic characters examined showed little intraspecific variation. Principal component analysis of the meristic variables was not informative.

Sheared PCA of the morphometric variables separated individuals from the Cumberland and Tennessee drainages into non-overlapping clusters, with most separation occurring along the sheared PC 2 axis (Figure 2). Examination of loadings indicates Big South Fork specimens have shorter soft dorsal and anal fins, a shorter anal fin base, a more posteriorly placed anal fin, and a more robust body (Table 5). In addition, a larger maximum size was attained by males from Big South Fork (58 mm SL) and Little River (56 mm SL; Etnier and Starnes 1993) than males from Copper Creek (48 mm SL; Jenkins and Burkhead 1994). Females attained a larger maximum size in Big South Fork (maximum 54 mm SL) than Copper Creek (45 mm SL, Jenkins and Burkhead 1994) and Little River (47 mm SL; Layman 1991).

#### DISCUSSION

#### Conservation Status

Etheostoma percnurum occupies a greater range in the Big South Fork than previously known. We have established the existence of the species over a 19-stream km reach at six

Table 4. Meristic counts displaying little intraspecific variation of 65 *Etheostoma percnurum* from four populations in the drainages of the Tennessee and Cumberland rivers, Virginia and Tennessee. Means are followed by ranges in parentheses.

Meristic	Copper Creek, VA n = 20	$\begin{array}{c} \text{Big South Fork,} \\ \text{TN} \\ \text{n} = 8 \end{array}$	Little Rock, TN n = 31	Citico Creek, TN n = 6
Dorsal spines	6.9 (6–8)	7	6.61 (6-7)	6.50 (6-7)
Dorsal rays	11.50 (11–13)	11.75 (11-12)	11.62 (11-13)	11.67 (11-12)
Pectoral rays	12.75 (12–13)	13	12.52 (12-13)	12.67 (12-14)
Pelvic rays	6	6	6	6
Anal spines	1.95 (1-2)	2	2	2
Anal rays	7.25 (6–8)	7.25 (7-8)	7.19 (7-8)	7.50 (7-8)
Principal caudal rays	17.85 (17–18)	17.25 (16-18)	17.87 (17–18)	16.33 (16-17)
Pored lateral-line scales	24.20 (17-28)	27.63 (25-30)	25.94 (23-30)	31.00 (29-34)
Scales above lateral line	6.80 (6-8)	6.63 (6-7)	6.48 (6-7)	6.00 (5-7)
Scales below lateral line	8.95 (7-10)	8.25 (7-9)	8.71 (8-10)	8.00 (7-9)
Scales around caudal peduncle	24.05 (22-27)	23.13 (22-25)	23.52 (21-27)	21.83 (20-25)
Interorbital pores	6.05 (6-7)	6	6	6
Preoperculomandibular pores	10.00 (9-11)	10.13 (10-11)	10.03 (9-12)	9.50 (9-10)
Total vertebrae	33.55 (33-35; n = 11)	33.63 (33-35)		
Precaudal vertebrae	13.82 (13-15; n = 11)	14.13 (13-15)		_
Caudal vertebrae	19.73 (19–20; $n = 11$ )	19.50 (18–20)		

sites in the Tennessee reach of Big South Fork (five of these not previously reported) and report it here for the first time from six sites in the Kentucky reach of Big South Fork. A recent survey by Shute et al. (unpublished data) located *E. percnurum* as far upstream as the mouth of Blevins Branch, Tennessee, expanding the known range in the Big South Fork to a 22-stream km reach. However, most Kentucky specimens are known only from a 3-km reach, and additional populations in Kentucky are unlikely to be present. The Big South Fork harbors the only known population of *E. percnurum* in the Cumberland River drainage.

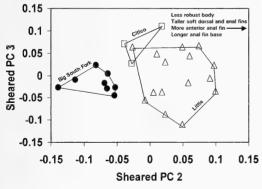


Figure 2. Morphometric scores on sheared PC axes 2 and 3 for 27 *Etheostoma percnurum* from Big South Fork of the Cumberland River, Little River, and Citico Creek, Tennessee.

The few other streams in this drainage that might harbor a relict population of this species have been well sampled (Burr and Warren 1986; Etnier and Starnes 1993). Our population estimate (300–600) over a 19-km stretch, though conservative, indicates considerably lower density in Big South Fork than Little River, Tennessee, where Layman (1991) estimated a population of 1023 E. percnurum in a 200-m reach in 1983. The highly restricted and localized distribution (mostly in about 3 stream km) of E. percnurum in Kentucky as well as its general rarity argue strongly for its immediate inclusion on the Kentucky state endangered/threatened species list as an endangered species.

The small distribution and population size of E. percnurum in Kentucky appears to be due to limited suitable habitat in Kentucky. Extensive alluvial streamside deposits (Pomerane 1964) are present from the mouth of Oil Well Branch to about 0.5 km above the mouth of Troublesome Creek, the reach with the largest Kentucky populations of E. percnurum. Similar alluvial deposits are almost entirely absent along the remainder of unimpounded portions of Big South Fork in Kentucky. Below the mouth of Bear Creek, suitable habitat continues to decline. Big South Fork narrows and becomes a series of long rapids strewn with large boulders, essentially lacking cobble and small boulder shoals. At Blue Heron, the

Table 5. Sheared principal component loadings for 27 morphometric variables for 27 *Etheostoma percnurum* from Big South Fork Cumberland River, Little River, and Citico Creek, Kentucky and Tennessee.

Measurement	Sheared PC 2	Sheared PC 3
Standard length	-0.005	-0.023
Head length	-0.115	0.067
Gape width	0.190	0.303
Pectoral fin length	0.103	0.222
Pelvic fin length	0.144	0.179
First dorsal fin height	-0.162	0.281
Second dorsal fin height	0.392	0.337
Anal fin height at third ray	0.228	0.367
Interorbital width	0.089	-0.099
Snout to occiput	-0.220	0.066
Snout to origin of pelvic fin	-0.044	0.010
First dorsal fin base length	0.143	0.034
Second dorsal fin base length	0.042	-0.161
Pelvic fin origin to anal fin origin	-0.344	0.068
Anal fin base length	0.334	-0.258
Second dorsal fin insertion to hypural	-0.037	0.118
Anal fin insertion to hypural	0.055	-0.290
First dorsal fin origin to anal fin origin	-0.133	-0.012
Pelvic fin origin to second dorsal fin origin	-0.054	0.097
Second dorsal fin origin to anal fin insertion	-0.178	-0.161
Anal fin origin to second dorsal fin insertion	0.296	-0.197
Occiput to pelvic fin origin	-0.025	-0.126
First dorsal fin origin to pelvic fin origin	-0.138	-0.045
Second dorsal fin origin to anal fin origin	-0.007	-0.108
Second dorsal fin insertion to anal fin insertion	0.110	-0.411
Head width	-0.271	0.022
Body width under second dorsal fin origin	-0.352	0.103

river widens, and some suitable habitat is present, although *E. percnurum* was not observed from this area. Below Blue Heron are the impounded waters of Lake Cumberland, certainly unsuitable for *E. percnurum*.

Although the mainstem of Big South Fork is protected from disturbances by the National Park Service, several tributaries (e.g., Bear Creek) are discharging low-quality because of mining in their watersheds. On 26 May 1998, following a rain the previous night, we observed extremely turbid water discharging from Bear Creek into the otherwise clear Big South Fork. Improvement in these impacted streams will help maintain the high-quality habitats in Big South Fork that are required by *E. percnurum* and other species (e.g., *Notropis* sp. "sawfin shiner" and *E. cinereum*) with restricted distributions in Kentucky.

We suggest periodic monitoring of the distribution and abundance of Kentucky *E. percnurum*. We consider both underwater observation and kick-seining around potential rock cover, our primary means of sampling in Big South Fork, as effective and non-lethal. We

recommend that electrofishing not be used to sample this rare species because of the potential harm it can do to fishes (Snyder 1995).

#### Natural History

Medium to large streams with silt-free rocky pools in the vicinity of riffles seem to be requirements for viable populations of *E. percnurum*. As pointed out by Etnier and Starnes (1993), the habitat of *E. percnurum* is essentially the same as that occupied by *E. cinereum*, a species we found almost invariably associated with *E. percnurum* in Big South Fork. Our general habitat description is similar to the habitat of *E. percnurum* in Little River (Etnier and Starnes 1993) and Copper Creek (Jenkins and Burkhead 1994).

Egg counts in Big South Fork (79–132; mean = 101) were higher than in Little River (23–200; mean = 79; Layman 1991). The higher egg counts per nest in Big South Fork may be attributed to the larger body size of those females. Using the equation of Layman (1991)  $\log C = -1.154 + 1.686 \log SL$ , females observed in this study would have 51–

61 mature ova, as compared to the 19-44 ova for the smaller females of Little River. Also, nest rock size typically was larger in Big South Fork (mean =  $24 \times 19$  cm) than in Little River (mean =  $16 \times 12$  cm; Layman 1991). Larger nest rock size in Big South Fork may simply reflect an abundance of larger nest substrata available. Alternately, this may be a behavioral adaptation in choosing more stable nest rocks in an area with high flows and prone to flash flooding. Additional studies are needed to explore these possibilities. Other nesting biology observations are generally consistent with those of Layman (1991) from Little River, Tennessee and Jenkins and Burkhead (1994) from Copper Creek, Virginia, except Jenkins and Burkhead reported nuptial males from moderate to swift runs.

The heavy infestations and high rate of infection of the black-spot disease observed in this study indicate that it may be an important source of mortality for E. percnurum. Although not previously documented for E. percnurum, the disease has been reported from many species of North American freshwater fishes. We observed belted kingfishers (Megaceryle alcyon) and snails, required intermediate hosts for the strigeid flukes that cause black-spot disease (Berra and Au 1978), to be common in Big South Fork. Heavy infestations have been reported to cause mortalities, particularly during the winter months, in Esox lucius, northern pike (Harrison and Hadley 1982), Lepomis macrochirus, bluegill (Lemly and Esch 1984), and Campostoma anomalum, central stoneroller (Ferrara and Cook 1998). Monitoring of the Big South Fork population of E. percnurum should include assessment of the extent of black-spot disease.

#### Systematics

The differences in squamation, body shape, maximum size, and nesting biology reported here indicate that Big South Fork populations certainly represent an independent evolutionary unit. Considering the relict distribution of this species it seems unlikely that any gene flow has occurred between Cumberland and Tennessee forms in thousands of generations. The morphological variation uncovered in this analysis supports the presence of deep phylogenetic partitions in *E. percnurum*. This pattern suggests that most of the overall diversity

is located among the populations rather than within populations (Meffe and Vrijenhoek 1988). Because the loss of even one of the remaining four populations of *E. percnurum* would cause a substantial loss in diversity of the species, conservation efforts should be directed to preserving as many populations as possible. Protection of the Big South Fork population seems particularly important in maintenance of diversity of *E. percnurum* because of the population's unique morphology and ecology.

#### **ACKNOWLEDGMENTS**

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#### **APPENDIX**

Specimens of *E. percnurum* used in morphological comparisons. Museum abbreviations follow Poss and Collette (1995). Parenthetical numbers after catalog numbers refer to the number of specimens used in the meristic and morphometric analyses, respectively.

Big South Fork of the Cumberland River. McCreary County, Kentucky: SIUC 24744 (1,1), SIUC 24761 (1,1), SIUC 24773 (5,5). Scott County, Tennessee: SIUC 24739 (1,1). Copper Creek. Scott County, Virginia: UMMZ 22038 (20,12). Little River. Blount County, Tennessee: UT 91.2719 (7,2), UT 91.2720 (9,4), UT 91.2721 (15,10). Citico Creek. Monroe County, Tennessee: UT 91.2558 (3,3), UT 91.4573 (3,0).

#### Scientists of Kentucky

#### David Wendel Yandell, M.D.

Every autumn when the banks along the Ohio blaze in color, the University of Louisville invites to the Falls City one of the nation's renowned surgeons as guest speaker for the annual Yandell Lecture.<sup>a</sup> The event celebrates the career of David Yandell (1826–1898), who taught clinical medicine and surgery at the school for more than 30 years. Because he cared passionately about improving medical education, Yandell led a relentless crusade to expand the knowledge of his professional colleagues and to upgrade the quality of care available to their patients.<sup>b</sup>

Born near Murfreesboro, Tennessee, Yandell spent his childhood in Lexington, where his father, Lunsford, taught chemistry at Transylvania University. In 1837 the elder Yandell and several of his colleagues founded the Louisville Medical Institute and the family moved to that city. The school attracted students from across the South and throughout the Ohio Valley because of its easy access and its fine faculty, which included Charles Caldwell, Jedediah Cobb, Daniel Drake, Samuel Gross, and Lunsford Yandell. By 1844 when David matriculated, the institute claimed an enrollment of 350.

Despite a growing number of medical schools throughout the nation, few 19th century physicians were well trained. Anyone could call himself a doctor and practice medicine. Most medical men learned their trade by serving a 2-year apprenticeship to a practitioner-preceptor. Some apprentices undoubtedly went along on house calls with their preceptors and thus learned to diagnose and medicate; others probably did little more than drive the doctor's buggy, attend his horse, and sweep out his office. Apprenticeships may have been inferior to medical schools, but no licensing board measured the knowledge of those that took advantage of either-or neither—program.

Throughout most of the century, medical schools had no entrance requirements. They were generally proprietary institutions plagued by feuding faculties, financial handicaps, competition for students, limited curricula, and inadequate teaching aides. To receive a degree, students were required to attend two 4-month sessions (an apprenticeship could be substituted for one session), pass an oral exam, and pay a graduation fee. The exam, apparently the only testing done by the faculty, could be a frightening experience. One of Yandell's classmates compared it to swallowing poison "with no stomach pump about, or [sleeping] with a man with smallpox." He described the examining professors as "dried up specimens of humanity who looked as if they had descended for the occasion from some anatomical museum and who have looked upon death, suffering and the annual ranks of medical aspirants" until their hearts were hard as

Although one of Yandell's professors labeled him a "damned unpromising specimen," he nevertheless passed the exam and graduated from the Louisville Medical Institute in March 1846.° A few weeks later he sailed to Europe to further his education in the schools of London and Paris. Many of his letters to his family appeared in the Western Journal of Medicine and Surgery and the Louisville Journal. The cocky Kentuckian greatly admired Sir Robert Lister's surgical techniques and described them in considerable detail, but he sneered at English students who were neither as "intelligent looking" nor as "fine in appearance" as his peers at home. He praised the politeness of French students and found the Parisian professors far more eloquent and interesting that their English brethren. Awed by the teaching hospitals, clinics, and internships available in Paris, Yandell claimed that in a single morning an industrious student could accompany professors through both the medical and surgical wards. And, if "fleet of limb he may follow Roux though his wards at the Hotel Dieu, Jobert through his at St. Louis and hear Velpeau lecture at La Charité!"

By the time he returned to Louisville in autumn 1848, Yandell enjoyed a reputation for excellence among both practitioners and lay-



Figure 1. David Wendel Yandell, ca. 1875.

men. His medications probably were no more successful than those of his colleagues, but his credentials and charm convinced many Louisvillians that he was very knowledgeable. Consequently, medical apprentices as well as patients flocked to him.

One of the problems facing would-be doctors was the lack of a practical method of

studying anatomy and surgical techniques. To aid them—and anyone else who desired the instruction-Yandell and a friend opened a dissection laboratory where they conducted classes in anatomy, physical diagnosis, and surgery. Following his marriage in summer 1851, Yandell closed the facility and moved briefly to Middle Tennessee. However, on his returned to Louisville in the mid 1850s, he opened a free outpatient clinic for Louisville's indigents. Modeled after the private clinics of Paris and financed by the fees of medical apprentices and university students, the dispensary supplemented rather than competed with university offerings. Consequently, it and its founder received considerable praise from local practitioners and from doctors who toured it during the 1859 state medical conclave in Louisville.

The success of the dispensary was partially responsible for Yandell being offered a position on the faculty, the realization of a decadelong dream. His major accomplishment as professor of clinical medicine was to talk the faculty into creating three "internships" for the university's top graduates. Unfortunately, the Civil War prevented implementation of the internships.

In summer 1861 Yandell resigned his professorship and received a commission in the Confederate medical department. He spent a few weeks with General Simon Bolivar Buckner's forces at Bowling Green, Kentucky, and then accepted the medical directorship of Albert Sidney Johnston's Army of the West, a command that extended from the Appalachian Mountains to Indian Territory. The task for this military monstrosity would have been a difficult one for a medic with many years of military experience and adequate resources; it was a gargantuan job for a novice. Epidemic diseases, shortage of medical supplies, inadequate hospital facilities, and poorly trained and inexperienced doctors plagued Johnston's army. To care for those felled by measles and other infectious diseases, respiratory ailments, and a variety of illnesses caused by bad food, contaminated water, and inadequate clothing, Yandell commandeered churches, public buildings, and vacant homes across southern Kentucky. When the number of ill exceeded space in the Kentucky facilities, he arranged with the L&N railroad to transport hundreds

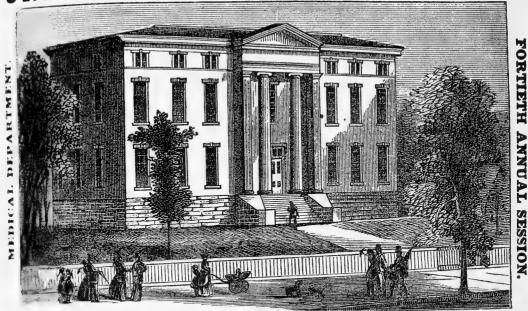
of them to the hospitals and convalescent centers he created in the Nashville area.

Yandell also supervised the employment of medical personnel. Regiments generally selected their own officers, including their regimental physician. Acknowledging the system's fallacy, Yandell created a examining board to screen all medical corps as well as civilian contract applicants. He headed the board and apparently was merciless in his questioning. When asked what he would do for a "shot right through there," the medical director pointing to his own knee, the victim of his grilling about wounds answered, "Well, sir, if it was you that was shot through there, I would not do a d[amne]d thing."

In mid February 1862 the Army of the West retreated into central Tennessee and then to the Corinth, Mississippi, area. In early April the army encountered the enemy near Shiloh Church, about 20 miles north of Corinth. More than 1700 Confederates died during the 2-day Battle of Shiloh; General Albert Sydney Johnston was among them, the victim of a torn popliteal artery. Earlier that morning Yandell had issued tourniquets to all of Johnston's staff, but no one thought to use the lifesaving device. With Johnston's death Yandell lost a friend as well as his coveted position in the Confederate medical department. The Army of the West was renamed and placed under the leadership of General P.G.T. Beauregard, who chose his own medical director. Yandell joined William J. Hardee's corps and traveled with the tactician's forces into Kentucky in autumn 1862. Following the battle of Perryville, he worked day and night administering to the wounded.

In spring 1863 Yandell was sent to Jackson, Mississippi, to "watch over" the health of General Joseph E. Johnston. The two men shared quarters, and the doctor often served as the general's aide, reading aloud his dispatches and writing the replies he dictated. Following the fall of Vicksburg, Yandell analyzed the sad state of affairs in Mississippi that culminated in the river town's loss; he laid the blame on officials in Richmond. His unwise comments reached Confederate president Jefferson Davis. Sensitive to criticism, Davis "banished" Yandell to the Trans Mississippi where the meddling doctor would have "less opportunity for exercising undue influence on the army

### UNIVERSITY OF LOUISVILLE.



CORNER OF EIGHTH AND CHESTNUT STREETS.

Figure 2. Medical Department, University of Louisville, Kentucky, ca. 1870.

and community." Yandell served the remainder of the war as medical director for the army of Edmund Kirby Smith.

Yandell labeled the Civil War a "great though terrible school." During his 4 years with the army he treated many medical and surgical problems and broadened his understanding of hospital management. He also made valuable contacts and won the admiration of physicians and laymen on both sides. The war's most profound effect, however, was to sharpen Yandell's awareness to the nation's large number of poorly trained and incompetent doctors. To the correction of this shortcoming, he dedicated the remainder of his life.

Returning to Louisville and his medical practice in July 1865, Yandell opened the Louisville Clinic, a large and efficiently organized facility; in 1869 he rejoined the University of Louisville's medical department. During the war the university had built a small dispensary and contracted two local doctors to operate it. Within a few weeks after Yandell rejoined the faculty, the contract doctors began to complain. They believed that his position on the

faculty gave Yandell's Louisville Clinic an "immense advantage" over the one they operated. Yandell offered to "divide the influence of his name and services equally between the two dispensaries, to lecture an equal number of times at each and receive no fee from either." His university colleagues refused his offer and passed a resolution that prohibited professors from teaching private classes. Yandell complained bitterly about the ex post facto ruling and refused to sever his connection with the Louisville clinic. Faculty pressure, however, forced him to rescind his decision.

Because the university's dispensary was too small to accommodate the students in small groups, much less for class instruction, Yandell began to push for an enlarged university clinic and adjoining amphitheater. Clinical teaching was the "alpha and omega" of a good education, he insisted. By studying medicine "in the laboratory, under the microscope, at the dissection-table, in the wards of the hospital, and in the dispensaries where patients are seen, examined and prescribed for . . . students

learn the diagnosis of disease as well as its treatment," he insisted.

When his colleagues whined that Yandell's building scheme would bankrupt them, he suggested that perhaps the university could convert its academic building, occupied rentfree by Male High School, into a hospital maintained by the city. Indigents could receive free care provided by the faculty and could be studied by students. The scheme would have created one of the first teaching hospitals in the nation. Yandell's colleagues liked the idea but unfortunately the city declined to consider it.

Disappoined but not discouraged, Yandell commenced talks with wealthy Louisvillian Shakespeare Caldwell and convinced him that a hospital was an appropriate memorial to his recently deceased wife. At this facility, Yandell argued, the university's faculty could provide medical and surgical services for impoverished patients and allow students to aid in treating patients and to gain bedside experience. The university offered land near the medical school for the facility. Caldwell agreed to the plan but vetoed the proposed site. Saints Mary and Elizabeth Hospital opened in 1874 but Yandell's plan proved unsuccessful. The distance of several miles between the school and hospital discouraged students from visiting it, and many of the professors could not or would not contribute time beyond their classroom responsibilities to care for patients and supervise students.

Undaunted, Yandell continued his nagging. He suggested that each professor make a contribution and that the faculty thus underwrite the construction of a clinic. The opposition was overwhelming. In frustration, Yandell submitted his resignation with a blistering attack on his colleagues, whom he accused of being disinterested in the quality of medical education. Undoubtedly a few of his harrassed colleagues welcomed Yandell's decision but others knew the university could ill afford to loose its most distinguished professor. Yandell edited the widely read American Practitioner, had been elected president of the American Medical Association in 1872, served as its representative to the 1881 International Conference in London, and was one of the founders of the American Surgical Association (and would be its president in 1889). Consequently, his colleagues promised that if he rescinded his resignation, they would reconsider his plea and try to finance the clinic. Yandell agreed. The university's treasurer cashed city bonds that the school had held for years and the faculty secured a loan for the remainder. H.P. McDonald Brothers received a contract to design and build the clinic and amphitheater. The facility for which Yandell crusaded for nearly 2 decades opened in 1888.

Yandell fought with equal zeal for entrance requirements, an expansion of the curriculum, and a lengthened school year. Unfortunately, competition for students was keen among the city's many medical schools (10 medical schools operated in Louisville during the postwar decades); changes that might increase overhead, raise tuition, or limit admission threatened enrollment and thus the salaries of the proprietary faculty. Yandell's demand for internships likewise met with resistance. The various hospitals were eager to implement the post-graduate positions but wished to make them available to all of the city's medical schools. After years of haggling on how the recipients should be chosen and who would supervise them, the university and the Kentucky Medical School agreed to base the selection on academic merit. Unfortunately, there were only a half-dozen positions for the several hundred graduates eager for the experience.

Yandell's campaign for a more rigorous medical curriculum transcended the commonwealth's borders. In his 1872 presidential address before the delegates to the American Medical Association in Philadelphia, he spoke about the shortcomings of the nation's educational system in general and medical education in particular.

... there are grave defects in the education of many of our students and many of our practitioners of medicine. Not a few of them, I am afraid, have a very slight acquaintance with grammar or physical geography and too many of them know little about etymology and are bad spellers. It is a pity that this is so and I should be glad to see a different state of things."

Answering colleagues who urged the addition of math and Greek to the medical school curriculum, Yandell championed the creation of better primary and secondary schools, not the

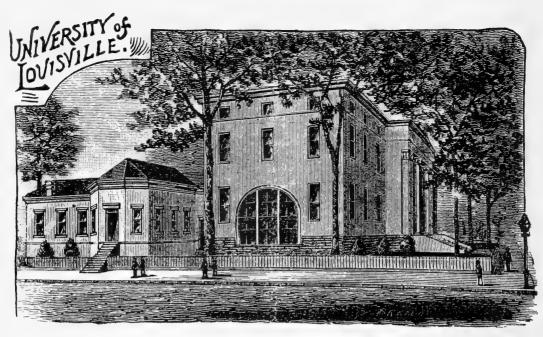


Figure 3. Medical Department, University of Louisville, Kentucky, ca. 1890. The wing on the left is the clinic for which David Yandell fought for 20 years.

teaching of "irrelevant topics" in medical school. Instead, he urged the extension of the lecture term, expansion of the requirements for graduation, increase in the number of professorships and course offering, and more instruction by demonstration. The graduates of the newly inaugurated system at Harvard, he stated, "may know less of Greek and mathematics, [but] are far better trained than they formerly were in clinical medicine and surgery and are better qualified to enter their duties as practitioners."

Although medical education began in the halls of academia, Yandell believed it should continue throughout a doctor's professional life. For 25 years he was the active senior editor of the *American Practitioner*, a monthly journal that had 2000 subscribers in 1876 and nearly 6000 by 1890. He filled the journal with original articles, reviews of American and European publications, synopses of clinical cases gleaned from other journals, and miscellaneous notes, editorials, and minutes of meetings of local, state, and national medical organizations. Yandell gave his readers a generous and well-balanced feast of medical knowledge and urged his readers to keep current

with developments in the profession. "Life is too short and science too long to permit time to be wasted," he frequently admonished.

As an classroom teacher Yandell had "few equals and no superiors," his students recalled, for he enriched their minds, made them wish to learn more, and flavored his lectures with bits of medical history and witty advice. A strong believer in human dignity, Yandell constantly reminded students that indigents deserved the same care and respect as the wealthy. He also urged these future doctors to stay abreast of and keep an open mind to new techniques but to use discretion in adopting them. "If one half of all the certain cures were but certain, the practice of medicine would be too simple to demand special study or require trained followers," he pointed out.

Yandell also advised that practitioners must know when to act with haste and when to proceed with deliberate caution. He delighted in telling about George IV, whose regular physician was too busy to attend to a small tumor on the royal derrière. Another doctor was called, the growth was removed, and the new doctor was knighted by his grateful sovereign. The reward might have been excessive, Yandell suggested, but a caring doctor never postponed "until the afternoon . . . messages left for you in the morning."

In their reminiscences, his former students lauded not only his teaching ability but also wrote of Yandell's surgical skills. He "cut to the line and to the required depth with geometric precission," one recalled. For demonstration purposes minor procedures were performed on hospital inmates and dispensary patients but for more difficult operations Yandell used cadavers. Most of the students sat 15 to 30 feet away yet claimed that their teacher's articulate and detailed explanations compensated for what they could not see. Student memoirs also recalled that Yandell always wore a freshly laundered coat in the "operating" room, scrubbed his hands and instruments with green soap before each procedure, and treated all incisions with compounds of iodine, bromine, and carbolic acid to prevent infection. He advised his students to do likewise. Prior to the general acceptance of antiseptic surgery, such cleanliness was unusual.

Yandell's admirers included personal friends and private patients, who recalled the bouguets he gave to shut-ins and the groceries he took to indigent families under the tactful guise that the patient needed special foods. Involved in community organizations, he served two terms on the city's school board, discussed history and literature with fellow members of the Filson and Salmagundi clubs, and founded the Louisville Kennel Club and Louisville Surgical Society. He counted among his close friends attorney Reuben Durrett, editor Henry Watterson, and former Confederates Basil Duke, William Preston, John B. Castleman, and Governor J. Proctor Knott, who appointed Yandell as surgeon general of the Kentucky militia. During one of his trips to Europe to attend a medical conclave, the British Medical Journal proclaimed that Yandell was a great favorite in local circles. The Medical Society of London elected him to honorary membership in 1883 and shortly thereafter the London Medical-Chirurgical Society named him an honorary fellow.

When an Englishman referred to him as a Yankee, Yandell protested, assuring that all of his blood "flowed through southern veins." The staunch Democrat served as one of Kentucky's hosts during the visits of three Republican presidents—Ulysses S. Grant, Rutherford B. Hayes, and Chester A. Arthur. At an official dinner in Louisville, Yandell entertained Grant with tales of his own Civil War escapades. While accompanying Arthur from Washington to Louisville, a newsman mistook the doctor for the president. Amused by the error, Yandell declared he was the "next best thing" to being president. He was the "Great Presidential Fetcher."

An articulate lecturer, talented surgeon, dedicated educator, and highly respected citizen, David Yandell was the most progressive and influential member of the university's medical faculty in the postwar era. In 1887 as part of the organization's celebration of its hundredth birthday, the College of Physicians and Surgeons of Philadelphia elected 10 American doctors to honorary membership. David Yandell was the only Kentuckian so honored. In spring 1895- the University of Louisville gratefully acknowledged his services when it placed "upon the brow of this our greatest son" the highest degree within its power, the degree of Doctor of Laws."

Nancy Disher Baird Kentucky Library Western Kentucky University Bowling Green, Kentucky 42101

#### **ENDNOTES**

- a. Further information about David Yandell is available in: Nancy Disher Baird, David Wendel Yandell, physician of old Louisville (Lexington: University Press of Kentucky, 1978). The major sources of information for the book-length biography and this article were the Yandell Family Papers (The Filson Club, Louisville); the Minutes of the Board of Trustees of the University of Louisville (University Archives); the Minutes of the Medical Faculty (Kornhauser Health Sciences Library, Louisville); General and Staff Officers' File, Old Army Section (National Archives, Washington); Andrew J. Foard Collection (Virginia State Library, Richmond); and Louisville's newspapers and Yandell's various published writings and speeches.
- b. Few Kentucky families have had greater influence on medical education than the Yandells of Louisville. Lunsford Pitts Yandell (1798–1878) taught chemistry and pharmacy at Transylvania's medical department, was one of the founders and teachers of the Louisville Medical Institute (renaned the Medical Department of the University of Louisville in 1846), edited two medical journals, and produced more than 100 sci-



Figure 4. Plaster bust of David Yandell by his niece Enid Yandell. Presented to the University of Louisville ca. 1925.

entific treatises relating to medicine and paleontology. His sons followed in his professional footsteps. William (1844–1901) became a well-known health officer in El Paso and led a campaign to improve sanitation and curb the spread of disease along the Texas-Mex-

ican border. Lunsford Jr. (1837–1885) taught medicine for nearly two decades at the University of Louisville. For additional information about Yandell's family members, see (1) Nancy Disher Baird, "A Kentucky Physician Examines Memphis, *Tennessee* 

- History Quarterly (autumn 1979), which concerns the early career of David's brother, Lunsford, Jr.; (2) "There Is No Sunday in the Army: The Civil War Letters of Lunsford Pitts Yandell, Jr.," The Filson Club History Quarterly (October 1980); (3) "Enid Yandell: Kentucky Sculptor," The Filson Club History Quarterly (January 1988). The best-known works by Enid (Lunsford Ir's. daughter) are the statue of Daniel Boone at the entrance to Louisville's Cherokee Park, Hogan's Fountain inside the park, and the statue of John Thomas in Nashville's Centennial Park. The Filson Club, Kentucky Historical Society, Georgetown College, Speed Museum, and Vanderbilt University (as well as scores of museums, parks, and private collections from Maine to Missouri) also own pieces of her works; and (4) Janet Brockmoller, "Doctor William Martin Yandell," Password, 21 (1976). Password is the quarterly publication of the El Paso County (Texas) Historical Society.
- c. In spring 1846 the state legislature created the University of Louisville. The "Academical Department" did not materialize until 1907 but the law department opened in autumn 1846 and the Louisville Medical Institute was incorporated as the university's medical department. The transaction for the latter was merely a legal form, for the faculty members continued to elect their own officers, choose new professors (whom the university's trustees automatically approved), collect student fees, and assess themselves when funds were needed for repairs and improvements on their building. In reality the medical school remained autonomous; only its name changed.
- d. In closing his presidential address, Yandell commented on admitting women to medical school. He could find no satisfactory reason, he said, why women might not succeed "in some line of our profession" for they were "able nurses," but he predicted that a female invasion of the traditional male domain would probably "end in no great results." Certainly he hoped that women would never embarrass the AMA by requesting membership. "I could not vote for that," he promised! Thirteen years later he addressed the graduation class of the short-lived Louisville School of Pharmacy for Women and gave a reason why he believed women might not be competent physicians or pharmacists: "Silence secures accuracy," and women were never quiet, he believed. Wishing the members of his audience good luck, he warned them against mixing a career and marriage, for "if you require your husbands to broil their own chops, you may expect them to wish at least to bray you in one of your own mortars."
- e. Yandell's last few years were marred by arteriosclerosis, which affected his memory and personality. In 1896 a stroke destroyed his remaining facilities. For nearly 2 years his family nursed the empty shell of a man who once charmed presidents and awed greenhorn medical students. Surrounded by those he loved best, David Yandell died 2 May 1898. Two days later a cortege of friends, colleagues, Confederate veterans, and a regiment of state militia conducted his remains to Louisville's Cave Hill Cemetery, where he was buried on a tree-dotted hillside overlooking a picturesque lake.

#### A Field Checklist of Kentucky Butterflies (Lepidoptera)

Charles V. Covell Jr.

University of Louisville, Louisville, Kentucky 40292-0001

On this and following page is the updated listing of all butterflies known from Kentucky. This supersedes the listing in Covell (1974), and consists of 144 butterfly species, 11 of which are either strays that rarely occur in the Commonwealth, or are known from old records and are not likely to be found in Kentucky now. In my opinion, the known resident and regularly transient butterfly fauna of Kentucky now stands at 133 species.  Scientific names follow Opler (1992), and English (common) names follow Glassberg (ed., 1992). Detailed records and remarks are available in Covell (1999).  I intend for this list to be photocopied by anyone wishing a pocket-sized list of Kentucky butterflies to use in the field. Anyone finding additions or suspected additions to this list is urged to contact me at the above address or at my e-mail address: covell@louisville.edu. I am sure there are a few other butterfly species native to or occasionally established temporarily in Kentucky.	<ul> <li>Urbanus proteus (Long-tailed Skipper) S</li> <li>Autochton cellus (Gold-banded Skipper) U</li> <li>Achalarus lyciades (Hoary Edge) U</li> <li>Thorybes bathyllus (Southern Cloudywing) C</li> <li>Thorybes pylades (Northern Cloudywing) C</li> <li>Thorybes confusis (Confused Cloudywing) U</li> <li>Staphylus hayhurstii (Hayhurst's Scallopwing) U</li> <li>Erynnis icelus (Dreamy Duskywing) C</li> <li>Erynnis brizo (Sleepy Duskywing) A</li> <li>Erynnis juvenalis (Juvenal's Duskywing) A</li> <li>Erynnis horatius (Horace's Duskywing) C</li> <li>Erynnis martialis (Mottled Duskywing) U</li> <li>Erynnis rarucco (Zarucco Duskywing) R</li> <li>Erynnis funeralis (Funereal Duskywing) R</li> <li>Erynnis baptisiae (Wild Indigo Duskywing) A</li> <li>Pyrgus centaurae (Grizzled Skipper) R</li> <li>Pyrgus communis (Common Checkered Skipper) C</li> <li>Pholisora catullus (Common Sootywing) C</li> <li>Nastrai lherminier (Swarthy Skipper) U</li> <li>Ancyloxipha numitor (Least Skipper) A</li> <li>Thymelicus lineola (European Skipper) F</li> <li>Hylephila phyleus (Fiery Skipper) F</li> <li>Hesperia leonardus (Leonard's Skipper) F</li> </ul>
ny in Kentucky.	
LITERATURE CITED	[Hesperia sassacus (Indian Skipper)] E
<ul> <li>Covell, C. V., Jr. 1974. A preliminary checklist of the butterflies of Kentucky. J. Lepid. Soc. 28:253–256.</li> <li>Covell, C. V., Jr. 1999. The butterflies and moths (Lepidoptera) of Kentucky: an annotated checklist. Kentucky State Nature Preserves Comm. Sci. Techn. Ser. 6.</li> <li>Glassberg, J. (ed). 1995. North American Butterfly Association (NABA) checklist and English names of North American butterflies. North American Butterfly Association, Morristown, NJ.</li> <li>Opler, P. A. 1998. A field guide to eastern butterflies. Houghton Mifflin, Boston, MA.</li> </ul>	<ul> <li>Polites peckius (Peck's Skipper) A</li> <li>Polites themistocles (Tawny-edged Skipper) A</li> <li>Polites origenes (Crossline Skipper) F</li> <li>Wallengrenia otho (Southern Broken-dash) S</li> <li>Wallengrenia egeremet (NorthernBroken-dash) A</li> <li>Pompeius verna (Little Glassywing) U</li> <li>Atalopedes campestris (Sachem) A</li> <li>Anatrytone logan (Delaware Skipper) U</li> <li>Poanes hobomok (Hobomok Skipper) F</li> <li>Poanes zabulon (Zabulon Skipper) C</li> <li>Poanes yehl (Yehl Skipper) C</li> <li>Poanes viator (Broad-winged Skipper) R</li> <li>Euphyes dion (Dion Skipper) F</li> </ul>
FIELD CHECKLIST OF KENTUCKY BUTTERFLIES	
by Charles V. Covell Jr., Dept. of Biology, Univ. of Louisville, Louisville, KY 40292-0001 Phone: (502) 852-5942.	<ul> <li>Amblyscirtes hegon (Pepper and Salt Skipper) U</li> <li>Amblyscirtes aesculapius (Lace-winged Roadside Skipper) U</li> </ul>
Locality	
Dates	— Amblyscirtes belli (Bell's Roadside Skipper) U- — Lerodea eufala (Eufala Skipper) R
Recorded by	——————————————————————————————————————
Epargyreus clarus (Silver-spotted Skipper) A	Battus philenor (Pipevine Swallowtail) A

—— [Battus polydamas (Polydamas Swallowtail)] S	Euptoieta claudia (Variegated Fritillary) C
——— Eurytides marcellus (Zebra Swallowtail) C	Speyeria diana (Diana Fritillary) C
——— Papilio polyxenes asterius (Black Swallowtail) C	Speyeria cybele (Great-spangled Fritillary) A
Papilio joanae (Joan's Swallowtail) R	Speyeria aphrodite (Aphrodite Fritillary) U
Papilio cresphontes (Giant Swallowtail) F	Speyeria idalia (Regal Fritillary) E
Papilio glaucus (Tiger Swallowtail) C	——— Boloria bellona (Meadow Fritillary) C
——————————————————————————————————————	[Boloria selene myrina (Silver-bordered Fritil-
——————————————————————————————————————	lary)] S
Pontia protodice (Checkered White) U	
——————————————————————————————————————	Charidryas nycteis (Silvery Checkerspot) C
Pieris rapae (Cabbage White) C	——————————————————————————————————————
——— Euchloe olympia (Olympia Marble) U	
Anthocharis midea (Falcate Orange Tip) C	
Colias philodice (Clouded Sulphur) A	——————————————————————————————————————
Colias eurytheme (Orange Sulphur) A	
	——————————————————————————————————————
——— Phoebis sennae (Cloudless Sulphur) C	——————————————————————————————————————
——— [ <i>Phoebis philea</i> (Orange-barred Sulphur)] S	
[Phoebis agarithe (Large Orange Sulphur)] S	toise Shell)] S
[Kricogonia lyside (Lyside Sulphur)] S	Nymphalis antiopa (Mourning Cloak) F
——— Eurema lisa (Little Yellow) F	
Nathalis iole (Dainty Sulphur) U	· ·
Feniseca tarquinius (Harvester) U	
— Lycaena phlaeas americana (American Copper)	
Lycaena hyllus (Bronze Copper) F	
— Atlides halesus (Great Purple Hairstreak) R	Limenitis arthemis arthemis (White Admiral)S
——————————————————————————————————————	
[Satyrium acadicum (Acadian Hairstreak)] S	ple) C
Satyrium edwardsii (Edwards' Hairstreak) C	Limenitis archippus (Viceroy) C (KY'S STATE
—— Satyrium calanus falacer (Banded Hairstreak) C	BUTTERFLY)
—— Satyrium caryaevorum (Hickory Hairstreak) U	——— Anaea andria (Goatweed Leafwing) U
—— Satyrium liparops (Striped Hairstreak) U	Asterocampa celtis (Hackberry Emperor) C
——————————————————————————————————————	
— Callophrys grynea (Juniper Hairstreak) C	— Enodia portlandia missarkae (Southern Pearly-
	Eye) U
——————————————————————————————————————	—— Enodia anthedon (Northern Pearly-Eye) C
Callophrys henrici (Henry's Elfin) C	
Callophrys niphon (Eastern Pine Elfin) U	Satyrodes appalachia (Appalachian Brown) C
——————————————————————————————————————	Cyllopsis gemma (Gemmed Satyr) U
——— Erora laetus (Early Hairstreak) R	— Hermeuptychia sosybius (Carolina Satyr) U
Calycopis cecrops (Red-banded Hairstreak) U	Megisto cymela (Little Wood Satyr) C
——————————————————————————————————————	——— Cercyonis pegala (Common Wood-Nymph) C
Leptotes marina (Marine Blue) R	——— Danaus plexippus (Monarch) C
— Everes comyntas (Eastern Tailed Blue) A	[Danaus gilippus (Queen)] S
	Additions:
Celastrina ebenina (Dusky Azure) C	Additions:
Celastrina neglectamajor (Appalachian Blue) U	
Glaucopsyche lygdamus (Silvery Blue) U	
——— Calephelis borealis (Northern Metalmark) U	[brackets indicate questionable specimens or sight records
Calephelis mutica (Swamp Metalmark) R	for Ky.]
— Libythaena carinenta bachmanii (American	A = Abundant; C = Common; F = Frequent; U = Un-
Snout Butterfly) C	common; R = Rare; E = probably endangered or extir-
Agraulis vanillae (Gulf Fritillary) U	pated; S = stray, not native to Kentucky.

# Notes on North American Elymus Species (Poaceae) with Paired Spikelets: I. E. macgregorii sp. nov. and E. glaucus ssp. mackenzii comb. nov.

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#### ABSTRACT

Elymus macgregorii R. Brooks & J.J.N. Campbell, sp. nov., is here described. Though widespread in eastern North America, it has been generally confused with *E. virginicus* and *E. glabriflorus*. It has a more open spike, with long awns, and there are other slight differences. It flowers about a month earlier and occurs in woodlands on relatively mesic, fertile soils. The new combination *E. glaucus* Buckley ssp. mackenzii (Bush) J.J.N. Campbell is provided for plants of rocky calcareous glades in the Ozark-Ouachita region, disjunct by 800 km from the main range of *E. glaucus* in western North America. Compared to other subspecies, ssp. mackenzii usually has narrower, pubescent leaf blades and longer glume awns.

#### INTRODUCTION

Elymus L. has been one of the most difficult genera of North American grasses to understand taxonomically. Published treatments range from simplistic (e.g., Gould 1975) to intricate (e.g., Bowden 1964). Elymus virginicus L. and closely allied taxa—a group characterized by relatively large, thick, basally indurate, bowed-out glumes that disarticulate from erect spikes—have been especially troublesome, although improvements in their treatment were advanced by the master's thesis of Brooks (1974). Variation within E. glaucus Buckley, a large complex species, also remains poorly understood (Snyder 1950, 1951; Stebbins 1957). I am currently completing a treatment of North American Elymus species with paired spikelets for the new Manual of North American Grasses (M.E. Barkworth, K.M. Kapels, and L.A. Vorobik, in preparation), which has already necessitated some taxonomic notes (Campbell 1995, 1996). This paper continues by (1) describing the following new species related to E. virginicus and (2) by providing a new subspecific combination in E. glaucus.

#### 1. ELYMUS MACGREGORII sp. nov.

In the late 20th century, it is rare to recognize a new species of vascular plant that is widespread in eastern North America. Nevertheless, based on much morphological and phenological study, Brooks (1974) showed that the plants described below are distinct. He initially treated them as *Elymus virgini* 

cus var. minor Vasey ex L.H. Dewey (1892, p. 550), but later (in McGregor et al. 1986, p. 1171) he noted that further study was needed. The type of E. virginicus var. minor has proven difficult to interpret: collected in "northern Texas" [without date] by [S.B.] Buckley s.n. (US 1020445), it consists of just one spike with an upper culm section and a few leaves. Its rachis internode lengths and awn lengths suggest that this specimen may be transitional between the species described below and *E. virginicus* var. *jejunus* (Ramaley) Bush. Moreover, "minor" cannot be used as a new specific epithet in this genus because it has already been used for a different species, as E. minor (J.G. Smith) M.E. Jones (1912), a name based on Sitanion minus J.G. Smith, which is now an accepted synonym of E. elymoides (Raf.) Swezey [=S. hystrix](Nutt.) J.G. Smith.].

### **Elymus macgregorii** R. Brooks & J.J.N. Campbell, sp. nov. Figure 1a.

Plantae caespitosae, plèrumque glaucopruinosae. Culmi 40–120 cm longi, erecti vel leviter decumbentes; nodi plerumque nudi. Foliorum vaginae glabrae vel raro villosae; ligulae minus quam 1 mm; auriculae 2–3 mm, purpurascentae vel nigrescentae; laminae 7–15 mm lata, laxae, supra glabrae vel interdum villosae, nitido-atrovirides sub pruina pallidoglauca. Spicae 4–12 cm longae, 2.5–4 cm latae, erectae, exsertae; nodi 9–18, unusquisque 2(3) spiculis; internodiae 4–7 mm longae, tenuiae (sectionibus angustissimis ca. 0.3

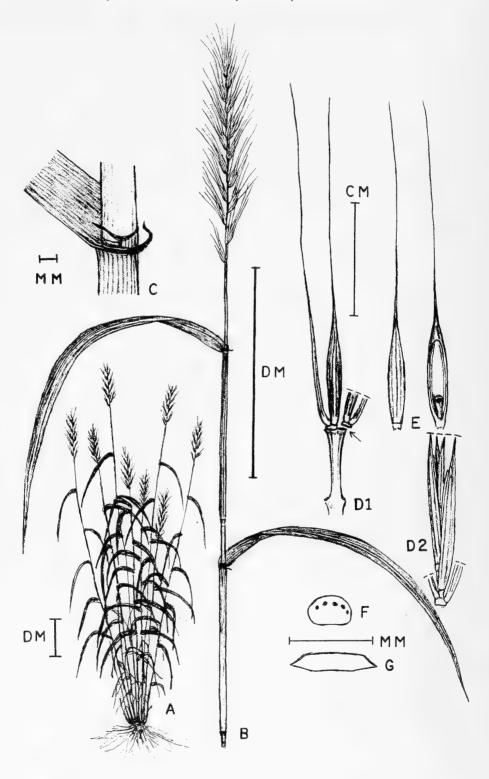
mm), sine angulis dorsalis prominentis. Spiculae 10-15 mm longae (minus aristae), effusae, glaucae vel maturite stramineae-fuscae, cum (2)3-4 flosculis, flosculus infirmus cadens cum glumae et basa rachillae affixae. Glumae 8-16 mm longae, 1-1.8 mm latae, basalia 1-3 mm induratae (nervis obscuris) et moderate exarcuatae, corpe lineari-lanceolato, plerumque glabro vel scabro, venis (2)4-5(8), arista 10–25 mm longa, stricta vel interdum contorta in spiculae infimae; lemmae 6-12 mm longae, plerumque glabrae vel scabrae vel interdum villosae ad hirsutae, arista (15)20–30 mm, stricta; paleae 6–10 mm longae, obtusae; antherae 2–4 mm longae, plerumque manifestae e mensis quintus serotinus ad mensis sextus medius; chromosomatum numerum. 2n = 28.

Plants cespitose, usually glaucous-pruinose. Culms 40-120 cm, erect or slightly decumbent; nodes mostly exposed. Leaf sheaths glabrous or rarely villous; ligules under 1 mm: auricles 2-3 mm, purplish to black; blades 7-15 mm wide, lax, glabrous or occasionally villous above, dark glossy green under the pale glaucous waxy bloom. Spikes 4-12 cm long, 2.5–4 cm wide including awns, erect, exserted; nodes 9-18, each with 2(3) spikelets; internodes 4-7 mm, thin (narrowest section ca. 0.3 mm), without prominent dorsal angles. Spikelets 10–15 mm (minus awns), spreading, glaucous then maturing to pale yellowish brown, with (2)3-4 florets, the lowest floret disarticulating with glumes and rachilla base attached. Glumes subequal, 8-16 mm long, 1-1.8 mm wide, the basal 1-3 mm indurate (with veins hidden) and moderately bowed out, the body linear-lanceolate, usually glabrous or scabrous, (2)4–5(8)-veined, the awn 10–25 mm, straight or occasionally contorted in the lowest spikelets; lemmas 6-12 mm, usually glabrous or scabrous, occasionally villous to hirsute, the awn (15)20-30 mm, straight; paleas 6-10 mm, obtuse; anthers 2-4 mm, usually evident from mid-May to mid-June. Chromosome number, 2n = 28 (Brooks 1974).

TYPE: U.S.A., KENTUCKY, Fayette Co., wooded banks of West Hickman Creek near Armstrong Mill Road, 31 May 1998, *J. Campbell 98-001* (HOLOTYPE: US; ISOTYPES: KY, KANU, KNK, MADI, MO, NCU, WIS). I have annotated many collections at

KANU, KNK, KY, ISC, MADI, MO, NCU, OKL, TEX, UARK, US, UTC, VDB, WIS, and elsewhere with the above name or with the earlier suggested names E. virginicus var. minor and "E. interior" ined. A distribution map is provided in Figure 1b, and recorded counties are listed in the Appendix. Elymus macgregorii occurs mostly in the Mississippi River and Ohio River drainages (Texas to South Dakota, Alabama to Ohio), but it also extends eastward to the Piedmont and New England (North Carolina to Maine), and westward to central Texas (Figure 1b). It has not yet been confirmed from Canada, but there is a possible atypical specimen from Nova Scotia (see Appendix), and the illustration of "Elymus hystrix L." in Dore and McNeill (1980) appears to be E. macgregorii, based on the long awns, distinct glumes, and upward-pointing spikelets. Its typical habitats are in mesic woodlands and thickets on fertile alluvium or, in a few regions (such as the "Bluegrass" of Kentucky, Indiana, and Ohio), on unusually fertile, base-rich upland residuum. In range and habitat, this species is somewhat similar to two widespread oaks (Little 1971), Quercus macrocarpa Michx. (excluding Q. mandanensis Rydb.) and Q. muhlenbergii Engelm. (excluding Q. prinoides Willd.).

Elymus macgregorii has been overlooked largely because its morphological distinctions are often not obvious at first inspection, especially in the herbarium. Yet, in 20 years of experience with this species in Kentucky, near the center of its range, I have found that, in addition to having some slight visible differences, it consistently flowers about a month earlier than its closest relatives, E. glabriflorus (Vasev ex L.H. Dewey) Scribn. & C.R. Ball and E. virginicus, including var. jejunus (Ramaley) Bush and var. intermedius (Vasey ex A. Gray) Bush. By recording dates of anthesis in the field and herbarium, I have confirmed the results of Brooks's (1974) garden studies that first demonstrated this phenological difference. I have also observed that the habitat of E. macgregorii is more restricted to woodlands on highly fertile, mesic soils, which, due to their productivity, are prone to much biotic disturbance. For example, this species appears to have been particularly abundant before settlement, along with the globally threatened



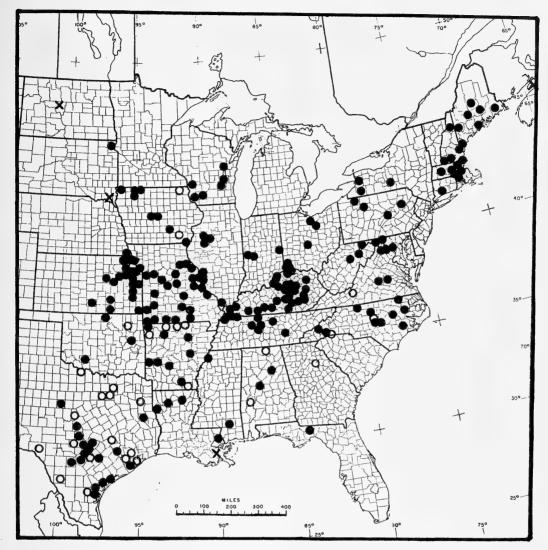


Figure 1b. Elymus macgregorii: mapped county records (eastern North America). Solid dots show counties recorded with typical plants; open dots show counties recorded with only atypical plants that may be transitional to E. virginicus var. jejunus (see text); crosses show counties with other atypical plants of uncertain identification, which may be hybrids in some cases. This map is based on herbarium specimens examined by the author (see Appendix) (a complete search of all major state herbaria has not yet been made).

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Figure 1a. Elymus macgregorii R. Brooks & J.J.N. Campbell.—A. Habit.—B. Upper portion of culm with mature spike, viewed on plane with alternating spread of spikelets.—C. Sheath summit and blade base.—D1. Mature rachis internode and glumes, viewed in plane of spikelet spread (with abaxial view of central glume in spikelet, and largely side view of lateral glume); arrow indicates disarticulation of right-hand glume pair and attached rachilla base from rachis.—D2. Spikelet, with lateral view of florets.—E. Mature floret in abaxial view (left) and adaxial view (right); note rounded palea summit shorter than lemma body.—F. Cross-section of mature, indurate glume base, showing embedded vascular bundles on abaxial side.—G. Cross-section of central rachis internode, showing only slight angles on abaxial side. Drawn from unpressed robust material in the same population as the holotype.

Trifolium stoloniferum Muhl., in the open, ungulate-browsed woodlands of Kentucky's Inner Bluegrass plains (Campbell et al. 1988). Such vegetation is now virtually all cleared for farmland, and E. macgregorii survives only in forested stream corridors, woodlots, and fencerows without frequent grazing or mow-

Elymus macgregorii has often been confused with E. glabriflorus [syn. E. virginicus var. glabriflorus (Vasey ex L.H. Dewey) Bush]. The latter is a species of the southeastern United States, typical of native grasslands and open woodlands on subhydric to subxeric sites that need not have particularly high soil fertility. Elymus macgregorii usually sheds pollen and begins to set seeds in mid-May to mid-June, as compared with mid-June to late July in E. glabriflorus. It has a more open spike with longer, more exposed rachis internodes ca. 4-7 mm, compared with 3-5 mm in E. glabriflorus. The spikes are often shorter and typically have about 9-18 nodes; in contrast, spikes of E. glabriforus typically have 15-30 nodes, unless unusually stunted. In many cases, there may be little or no obvious vegetative differences. However, in fresh condition the leaves of E. macgregorii are generally lax and dark, glossy green under the distinct, pale, glaucous, waxy bloom, whereas those of E. glabriflorus range from lax (especially in shade) to ascending and somewhat involute (especially in sun), and they are generally paler, duller green with or without a waxy bloom. The auricles of *E. macgregorii* are typically prominent, ca. 2–3 mm, and turn from purplish to black at maturity, whereas those of E. glabriflorus (and E. virginicus) are typically less developed, ca. 0-2 mm, and only brownish at maturity.

Both Elymus macgregorii and E. glabriflorus can be distinguished from E. virginicus by their fully exserted, wide spikes (ca. 2.5–6 cm wide including awns), with more spreading spikelets and longer lemma awns (ca. 15–40 mm). Spikes of E. virginicus vary from fully exserted to remaining partly enclosed by the upper leaf sheaths and are only 0.7-2 cm wide; its spikelets are appressed to slightly spreading, and lemma awns are 1–15(20) mm long. While no consistent vegetative differences have been found, E. macgregorii and E. glabriflorus are sometimes pubescent in spikes

(especially lemmas) and leaves (sheaths and upper blade surfaces), whereas E. virginicus is generally glabrous to scabridulous except for a few varieties or forms that have rather narrow ranges in distribution or habitat. A later paper in this series will present a detailed key to these species and other Elymus spp. with

paired spikelets.

Variation within Elymus macgregorii deserves further study. Throughout much of its range, plants with pubescent spikes (especially lemmas) occur at scattered locations, but these have not yet been reported in distinct habitats or in large enough populations to warrant taxonomic recognition. However, some plants from Missouri, Arkansas, Oklahoma, and Texas, including the type of E. virginicus var. minor, do deserve more detailed study for possible recognition. These have smaller spike dimensions (with internodes down to 3-4 mm and awns down to 10 mm), and often less glaucous foliage. They may represent a transition to E. virginicus var. jejunus, though often with distinctly villous leaves. Thus, separation of E. macgregorii from E. virginicus var. jejunus may remain difficult in this region without further research.

My examination of herbarium material suggests that Elymus macgregorii forms rare natural hybrids with E. virginicus, E. glabriflorus, E. hystrix L., and perhaps E. villosus Muhl. ex Willd. Also, while misidentified as E. virginicus var. intermedius (Vasev ex A. Gray) Bush, E. macgregorii appears to have been artificially hybridized by Stebbins amd Snyder (1956) with E. glaucus, E. stebbinsii Gould, and Pseudoroegneria spicata (Pursh) A. Löve [syn. Agropyron spicatum Pursh]. This misidentification is indicated by examination of their probable voucher material at US (G.L. Church s.n., Limington, Vermont), and by its attributed characters in their Table 2: rachis internodes ca. 7 mm, glumes ca. 28 mm, and lemmas ca. 28 mm (including awns).

Elymus macgregorii is named in honor of the Clan M[a]cGregor, as represented by two descendants: (1) Ronald Leighton McGregor, emeritus professor and former herbarium director at the University of Kansas (Lawrence), Great Plains botanist (McGregor et al. 1986), and supervisor of Brooks (1974); and (2) John MacGregor (of Nicholasville, Kentucky), an outstanding Kentucky naturalist, explorer of

Inner Bluegrass thickets and other disturbed places, and my occasional collaborator (e.g., Campbell et al. 1994). An appropriate English name is "early wild-rye" since this is the first species of *Elymus* to flower in east-central North America.

### 2. ELYMUS GLAUCUS ssp. MACKENZII comb. nov.

Elymus glaucus ssp. glaucus is a widespread taxon in western North America (Figure 2b). There are two other described subspecies, which are confined to regions along the Pacific Coast and adjacent mountain ranges, and there is much additional variation that deserves further attention (Snyder 1950, 1951; Stebbins 1957). For example, some plants can have solitary spikelets at most nodes, often resembling E. trachycaulus (Link) Gould ex Shinners according to M.E. Barkworth (Utah State University, pers. comm., June 1997). The main range of *Elymus glaucus* extends east to the western Great Plains, but this species is unknown or very rare in most of Texas, Oklahoma, Kansas, and Nebraska. Its northern range extends east to a few scattered records in the Great Lakes region (e.g., Voss 1972). Some Great Lakes records, however, are questionable due to possible confusion with E. trachycaulus, or possible post-Columbian spread of E. glaucus to the east.

This section of the paper is concerned with the remarkably disjunct southeastern segregate of this species in the Ozark Mountains and Ouachita Mountains of eastern Oklahoma, northwestern Arkansas, and southwestern Missouri. These plants occur mostly in rocky, calcareous, xeric openings. They were originally described as Elymus mackenzii Bush (1926), based on Missouri specimens, but no subsequent author has recognized them as distinct from E. glaucus. The taxon is resurrected here as a subspecies of E. glaucus because of its slight-morphological differences and its great disjunction of about 800 km from the nearest records of E. glaucus to the west and north.

**Elymus glaucus** Buckley ssp. **mackenzii** (Bush) J.J.N. Campbell, comb. nov. Figure 2a.

Basionym: *Elymus mackenzii* Bush, Am. Midl. Naturalist 10:53, 1926.

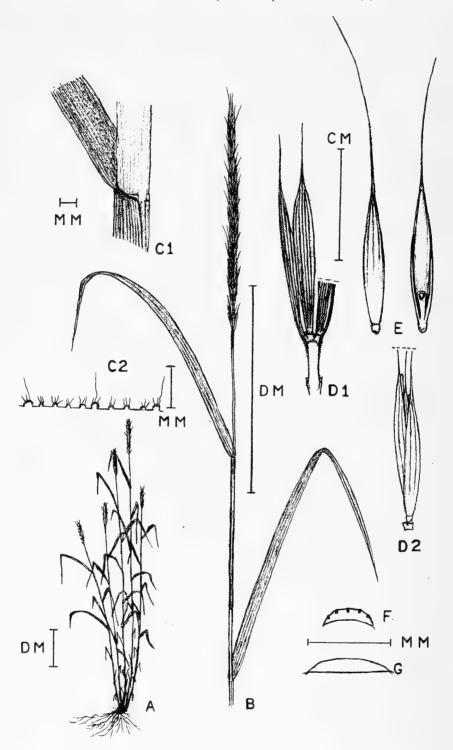
TYPE: U.S.A., MISSOURI, [Barry Co.] Eagle Rock, 15 Jun 1897, B.F. Bush 77 (HOLOTYPE: US 318128!).

OTHER COLLECTIONS EXAMINED. U.S.A., ARKANSAS: Carroll Co., wooded northeast facing slope along White River at Catron Bend, 5 miles northwest of Eureka Springs, 27 Jul 1953, M.J. May 12 (UARK); Polk Co., Rich Mountain, margin of rich woods, 30 Jun 1967, G.E. Tucker 5439 (APCR, NCU). MISSOURI: Barry Co., 3 miles east of Roaring Rv. State Park on high limestone Juniperus glade, 10 miles southeast of Cassville, H.H. Iltis et al. 1, 3 Jul 1960 (WIS, KY). OKLAHOMA: Le Flore Co., summit of Rich Mt. near Arkansas line, damp ledge, rock in full sun, 24 Jun 1980, R. Kral 65492 (VDB).

Another Missouri record is from Moore (1954), who listed "Elymus mackenzii" from Stone Co. on xeric limestone bluffs along the White River. I have not yet located voucher collections by Moore, but H.H. Iltis (Univ. of Wisconsin, pers. comm., Nov 1996) worked with him and has confirmed the identification. Also, Stevermark (1963) mapped E. glaucus from eight counties in southwestern Missouri (Barry, Barton, Jasper, Lawrence, Newton, Ozark, Stone, and Taney). His collections, mostly at UMO (and perhaps F), have been partly examined by Yatskievych (1999), whose description of *E. glaucus* suggests that all Missouri plants belong to ssp. mackenzii, except perhaps for a old collection from Jackson Co. with unknown location and affinity: 21 Jul 1892, B.F. Bush 2849 (UMO). Stevermark (1963) noted that the species is "usually found on rocky limestone ledges of bluffs along White River and tributaries and other streams."

The following key distinguishes the described subspecies of *Elymus glaucus*. However, although ssp. *jepsonii* was recognized in Hickman (1993), this taxon may not deserve subspecies status according to M.E. Barkworth (pers. comm., April 1999). A complete revision of this complex species is needed.

Lemma awns (5)10-25(35) mm; glume awns (0.5)1-8(9) mm; blades glabrous or variously pubescent.



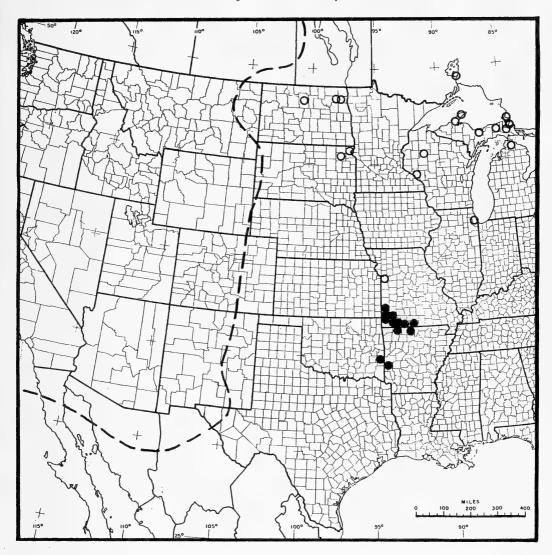


Figure 2b. Elymus glaucus: mapped county records of ssp. mackenzii and outlying eastern records referable to ssp. glaucus (central North America). Solid dots show ssp. mackenzii (see text for collection details); open dots show other eastern records of E. glaucus, including plants that are similar to E. trachycaulus, based on authorities (e.g., Bowden 1954; Gould 1975; McGregor et al. 1986; Voss 1972), and on scattered collections seen by the author. The dashed line shows approximate eastern boundary of main range of E. glaucus based on reliable data accumulated by M. E. Barkworth (pers. comm.); there are no other records between this line and ssp. mackenzii.

 $\leftarrow$ 

Figure 2a. Elymus glaucus ssp. mackenzii (Bush) J.J.N. Campbell.—A. Habit.—B. Upper portion of culm with mature spike, viewed on plane with alternating spread of spikelets.—C1. Sheath summit and blade base.—C2. Adaxial blade pubescence.—D1. Mature rachis internode and glumes, viewed in plane of spikelet spread (with abaxial view of central glume of spikelet, and largely side view of lateral glume); note lack of prompt disarticulation by rachilla base from rachis.—D2. Spikelet, with lateral view of florets.—E. Mature floret in abaxial view (left) and adaxial view (right); note narrowly truncate palea summit almost equalling lemma body.—F. Cross-section of mature glume base, showing surficial vascular bundles on abaxial side.—G. Cross-section of central rachis internode, showing lack of angles on abaxial side. Drawn from robust material of H.H. Iltis et al. 1 (WIS, KY).

- Blades usually 4–13 mm wide, glabrous to strigose above, or occasionally pilose to hirsute with hairs of fairly uniform length; glume awns 1–5 mm
  - 3. Blades strigose, pilose, or hirsute; lemmas awns to 20 mm.
  - ssp. jepsonii (Burtt Davy) Gould
    Blades glabrous, scabrous, or sparsely strigose; lemma awns to 35 mm ...... ssp. glaucus

ssp. mackenzii (Bush) J.J.N. Campbell

Very few vascular plants are known to have such striking disjunctions from western ranges to the Ozarks or Ouachitas, as is the case in *Elymus glaucus. Mimulus floribundus* Douglas ex Lindl. (Scrophulariaceae) is a widespread species of the Rocky Mountains that occurs on wet dripping cliffs in the Ozark Mountains of Arkansas (Moore 1959; Smith 1991). Like *E. glaucus*, the eastern plants of *M. floribundus* appear to have some morphological distinction, but in this case a subspecific epithet has not yet been published (H.H. Iltis, pers. comm., April 1999).

#### **ACKNOWLEDGMENTS**

For their assistance and encouragement, I am particularly grateful to Mary Barkworth, Ralph Brooks, Hugh Iltis, Dan Nicholson, Max Medley, John and Charlotte Reeder, Rob Soreng, and George Yatskievych. I thank the curators of the various herbaria where I have studied material or obtained loans.

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#### APPENDIX: COUNTIES AND HERBARIUM SOURCES FOR SPECIMENS OF *ELYMUS MACGREGORII* ANNOTATED BY THE AUTHOR.

I have annotated collections as *Elymus macgregorii* (or *E. virginicus* var. *minor*) from the following counties. Herbarium acronyms (Holmgren et al. 1990) are in parentheses. Uncertain identifications due to incomplete or depauperate specimens or probable hybrids are generally excluded unless they represent possible county records (with caveats noted in parentheses). However, a frequent, mostly southwestern, variant that may be transitional to *E. virginicus* var. *jejunus* is included and shown by asterisks.

CANADA. NOVA SCOTIA: "Bass River" (MADI, US)? [30 Jul 1875, J. Fowler s.n.—this northernmost specimen is atypical and might be at least transitional to E. virginicus, but is included here as the only possible record seen so far from Canada].

U.S.A. ALABAMA: Bibb (VDB); St. Clair (NCU). ARKANSAS: Ashley (UARK\*); Baxter (UARK\*); Benton (MADI, NCU,\* UARK\*); Boone (KANU\*); Bradley (VDB); Fulton (UARK\*); Hot Spring (US); Independence (KANU); Montgomery (VDB); Newton (BE-REA\*, UARK\*); Polk (ISC, VDB); Prairie (ISC): Washington (NCU,\* UARK,\* US\*). CONNECTICUT: New Haven (US). DIS-TRICT OF COLUMBIA (ISC, MADI). FLORIDA: Leon (TEX) [Godfrey 84158, "common in ditches"-could this be adventive?]. GEORGIA: Clarke (US\*). ILLINOIS: Fulton (US); Jersey (KNK); Knox (MO); Peoria (US); Union (MADI, KNK). INDIANA: Parke (US); Putnam (US); Wayne (US). IOWA: Boone (ISC); Clay (ISC, MO\*); Dickinson (ISC); Emmet (ISC); Jefferson (ISC\*); Lee (ISC); Lyon (ISC); Mahaska (ISC); Story (MO,\* NCU\*); Winneshiek (ISC\*). KANSAS: Allen (KANU); Anderson (KANU); Atchison (KANU\*); Brown (KANU); Butler (KANU); Cherokee (KANU); Cowley (KANU, MADI); Dickinson (KANU); Doniphan (KANU\*); Douglas (KANU); Franklin (KANU); Jackson (KANU); Jefferson (KANU); Kingman (KANU); Leavenworth (KANU); Miami

(KANU); Morris (KANU); Nemaha (KANU); Shawnee (KANU): Wabaunsee (KANU): Wvandotte (KANU). KENTUCKY: Anderson (EKY); Barren (KY); Boone (KNK); Calloway (NCU); Campbell (KNK, NCU); Casey (BE-REA, KY); Christian (VDB); Clark (KY); Clay (MO); Fayette (KY, US); Franklin (KY); Fulton (KY, VDB); Grant (KNK); Graves (VDB); Green (EKY); Hickman (KY); Jefferson (KY, NCU, TEX, UTC); Jessamine (KY); Kenton (KNK, VDB); Laurel (BEREA); Livingston (KY); McCracken (KY); Madison (KY); Oldham (KY); Owsley (KY); Pendleton (NCU); Pulaski (KY); Rowan (KY); Spencer (KY); Trimble (KY); Warren (WKY); Wayne (MORE); Wolfe (KY); Woodford (KY). LOU-ISIANA: Bienville (US); De Soto (US); Jefferson? (VDB—perhaps transitional to E. glabriflorus); Ouachita (NCU); St. Tammany (ISC). MAINE: Knox (NCU); Oxford (GH); Penobscot (ISC); Piscataquis (US); Washington (NCU); York (NCU, US). MARYLAND: Garrett (US); Montgomery (TEX); Washington (KANU). MASSACHUSETTS: Essex (US); Hampshire (MADI); Worcester (MADI, TEX). MISSISSIPPI: Forrest (US); Tunica (NCU). MISSOURI: Barry (MADI\*); Boone (MO); Christian (ISC\*); Cole (MO); Dade (MO, US); Franklin (MO); Greene (NCU); Jackson (ARIZ, ISC, MO, US); Jasper (MO, US); Jefferson (KANU, MO, NCU, UTC\*); Lafayette (MO); McDonald (MO); Monroe (MO); Morgan (MO); Oregon (MO); Pettis (MO); Phelps (MO); Pike (MO); Platte (MO); Ralls (MO, US, UTC); St. Charles (MO, UTC); St. Clair (TENN); Ste. Genevieve (MO); St. Louis (ISC, MO); Shannon (MO); Taney (MO); Texas (MO); Warren (MO). NE-BRASKA: Richardson (KANU, US\*). NEW HAMPSHIRE: Cheshire (GH); Coos (ISC, TEX, US); Hillsborough (GH, MADI, MO, TENN); Strafford (MADI). NEW YORK: Cattaraugus (MO); Tompkins (MO). NORTH CAROLINA: Alamance (US); Chatham (NCU\*); Harnett (NCU); Haywood (NCU); Henderson (NCU); Jones (NCU); Lee (NCU); Martin (NCU); Northampton (NCU); Polk (NCU\*); Stokes (NCU); Wilson (NCU). NORTH DAKOTA: McClean? (KANU—at least transitional to E. virginicus). OHIO: Ashland (TENN); Franklin (VDB); Hamilton (US); Huron (NCU). OKLAHOMA: Comanche (US); Kay (KANU); Muskogee (US); Rogers (VDB\*). PENNSYLVANIA: Elk (ISC); Luzerne (TEX); Snyder (TEX); Warren (ISC). RHODE ISLAND: Providence (GH). SOUTH DAKOTA: Clay? (MO—perhaps transitional to *E. virginicus*); Roberts (ISC). TENNESSEE: Bledsoe (VDB); Davidson (KANU, MADI, US, VDB); Dickson (VDB); Smith (VDB); Williamson (VDB). TEXAS: Austin (MO,\* TEX, US); Bexar (MADI,\* MO,\* TEX); Blanco (KANU, TEX); Brazoria (TEX); Brazos (TEX\*); Brown (TEX\*); Burleson (US\*); Collin (MO\*); Comal (MO, TEX,\* US\*); Dallas (MO\*); Dimmit (US\*); Fort Bend (TEX\*); Galveston (TEX\*); Gillespie (TEX\*); Gonzales (TEX); Harris (TEX\*);

Hays (TEX); Jim Wells (US\*); Llano (TEX); Medina (TEX); Nacogdoches (US); Nueches (MO); Refugio (TEX); San Patricio (ISC, MO, TEX); San Saba (MO); Tarrant (TEX,\* US\*); Taylor (ISC,\* MO, TEX); Travis (ISC\*, MO,\* TEX); Val Verde (TEX\*); Wichita (TEX\*). VERMONT: Essex (US). VIRGINIA: Buckingham (NCU); Clarke (NCU); Loudoun (NCU); Montgomery (NCU\*); Powhatan (NCU). WEST VIRGINIA: Jefferson (NCU); Morgan (NCU); Tucker (TEX); Upshur (NCU). WISCONSIN: Dodge (MADI); Fond Du Lac (MADI); Grant (MADI); Iowa (MADI); Outagamie (MADI); Vernon (MADI).

# Proterometra macrostoma (Digenea: Azygiidae): Distome Emergence From the Cercarial Tail and Subsequent Development in the Definitive Host

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#### ABSTRACT

The objectives of our study were (1) to evaluate effects of pH and pepsin on emergence of the *Proterometra macrostoma* distome body from its cercarial tail and (2) to examine morphological changes associated with maturation of this worm in the sunfish definitive host. Distome emergence from the cercarial tail was significantly faster at low pH (1.5–2.5). Addition of 0.5% pepsin appeared to accelerate this process. The number and maximum size of eggs increased in adult worms over the initial 18 and 24 days, respectively, in experimentally infected bluegill, *Lepomis macrochirus*. No other trends for change in worm size were noted. Mature eggs containing miracidia were first observed by day 18 postinfection.

#### INTRODUCTION

Proterometra macrostoma is a digenetic trematode widely distributed in eastern United States. The complete life cycle was first described by Horsfall (1933, 1934). The adult worm is found in the esophagus and stomach of sunfishes (Family Centrarchidae). Eggs containing fully developed miracidia are released into water with fecal material and are subsequently ingested by snails in the genus Elimia. Intramolluscan stages include sporocysts, rediae, and cercariae. During emergence from the snail, the distome body of the cercaria enters a vesicle/cavity within the cercarial tail and detaches from the latter structure. After it is released into the water, the cercaria's swimming behavior and large sizetailstem 4-8.9 mm long (LaBeau and Peters 1995)—make it attractive to potential definitive hosts, which rapidly ingest the worm. The progenetic (i.e., containing in uterus eggs in early cleavage) distome body is then liberated from the cercarial tail in the fish stomach and matures directly into an adult worm in the host stomach or esophagus.

The time frame and conditions that promote the emergence of the distome body from the cercarial tail have not been quantified. Horsfall (1934) reported emergence of *P. macrostoma* from the cercarial tail in a 1.0% solution of HCl, but it was unclear what pH, temperature, and time frame were used. Similarly, the subsequent development of this "liberated" distome in the definitive host has been based on morphological comparisons between

the distome body within the cercaria vs. the adult worm (Horsfall 1934). The time frame for changes associated with maturation of the adult worm in sunfishes has not been previously described.

The objectives of our study were (1) to evaluate the effect of pH and pepsin on emergence of the distome body from the cercarial tail of *P. macrostoma* and (2) to establish a time line for possible morphological changes during maturation in sunfish definitive hosts.

## **METHODS**

#### General Methods

Snails were collected in June and July 1998 from North Elkhorn Creek (38° 11′ 00″ N, 84° 29' 19" W) in Scott County, Kentucky. Snails were maintained under a protocol similar to that described by Riley and Uglem (1995). They were placed in white enamel pans filled with filtered creek water, held at 20-25°C under continuous light, and fed lettuce ad libitum. The water in the pans was changed every 2 days. When cercariae were required for experiments, any previously emerged worms were removed from these pans. The snail cultures were then placed in an environmental chamber in the dark at 20°C, which promoted a copious release of new cercariae within 2 hours.

# Distome Emergence

To assess the effect of pH and pepsin on distome emergence from the cercarial tail, a Ringer's solution for cold-blooded vertebrates (i.e., 6.5 g/liter NaCl, 0.05 g/liter KCl, 0.16 g/ liter CaCl<sub>2</sub> × 2H<sub>2</sub>0, 0.39 g/liter MgSO<sub>4</sub> × 7H<sub>2</sub>0, and 0.2 g/liter NaHCO<sub>3</sub>) was acidified with concentrated HCl to obtain pHs of 1.5, 2.0, 2.5, 3.0, 3.5, and 4.0. These values were within the physiological range (i.e., pH 1-5) of bluegill gastric juice reported by Norris et al. (1973). For the pH + pepsin experiments, 0.5% pepsin (Sigma: 1:2500; activity 600–1800 units per mg protein) solutions were made with each of the acidified salines. Cercariae less than 2.5 hours postemergence were used in these experiments. They were pipetted into individual beakers containing a particular precooled (20°C) acidified saline or acidified saline + pepsin, incubated at 20°C, and checked for complete distome emergence from the cercarial tail every 5 minutes for 1 hour. Each experiment consisted of six treatments and 60 worms, with 10 cercariae/treatment (=pH). Both the pH and pH + pepsin experiments were repeated three times.

A mean  $\pm$  SE (% distome emergence) was calculated for each time period and pH based on the three replicate treatments. A chisquare goodness-of-fit test was used to determine if significant differences in the number of emerging distomes existed due to pH at 5 and 30 minutes for both the acidified saline and acidified saline + pepsin experiments. A 2  $\times$  2 chi-square contingency test was used to assess differences in numbers of digeneans emerging from cercarial tails in acidified saline vs. acidified saline + pepsin at 5 and 30 minutes for each pH level assessed. A probability of P < 0.05 was considered significant for all

statistical tests.

#### Fish Infections

Young, hatchery-reared (Ken Jacobs, Bowling Green, KY) bluegill, Lepomis macrochirus (mean fork length = 5.2 cm), were used in this experiment. Fish were maintained in aerated 10 and 15 liter aquariums at 24.6°C and fed TetraMin (Tetra Sales, Blacksburg, VA). Prior to infection, one bluegill was placed into a 3.3 liter tank. After a 1–3 minute acclimation period, two cercariae were released into this tank. Visual confirmation of cercariae ingestion was made, and then the exposed fish was placed into a new aquarium. In addition, several bluegill were exposed to cercariae in mass

and subsequently maintained in the manner described above.

Every 3 days, beginning at day 0 (at 5 and 15 minute postinfection [PI]) and ending on day 24 PI, 3-5 fish were sacrificed and necropsied. Worms were removed from the stomach, placed on a slide under a coverslip, and examined with a compound microscope. The number and developmental stage of eggs were recorded. Developmental stages were based on the descriptions of Horsfall (1934): (1) Stage I—eggs containing a clear mass at their anterior end and a large vitelline mass at the posterior end, (2) Stage II—the vitelline mass in eggs is less apparent and signs of advanced cleavage are more obvious, and (3) Stage III further reduction in the vitelline mass, darkening of the egg into a yellow color, and appearance of bristle plates associated with the developing miracidium at the opercular end. The worm was then fixed and flattened under a coverslip with FAA (formalin-alcohol-acetic acid). Flukes were stained with Semichon's carmine, and permanent slides were made. Length and width measurements of worm size, oral sucker, acetabulum, pharynx, testes, and eggs were obtained from fixed/stained specimens with an ocular micrometer. Means ± SE in micrometers were calculated from these measurements for five time-intervals.

## **RESULTS**

The number of emergent distomes in the six acidified salines without 0.5% pepsin (Figure 1) was significantly different at 5 minutes ( $\chi^2 = 24.5714$ ; df = 5) and 30 minutes ( $X^2 = 42.4286$ ; df = 5). Similarly, the number of emerging digeneans in the six acidified salines with 0.5% pepsin (Figure 1) was significantly different at 5 minutes ( $\chi^2 = 53.3019$ ; df = 5) and 30 minutes ( $\chi^2 = 65.9895$ ; df = 5). The overall trend suggested that emergence of *P. macrostoma* from the cercarial tail was facilitated at lower pH levels (i.e., 1.5–2.5; Figure 1).

Significant differences were noted in emergence of digeneans from the cercarial tail when the effect of acidified salines was compared to acidified salines + 0.5% pepsin at 5 minutes (pH 1.5— $\chi^2$  = 20.3175, df = 1; pH 2.0— $\chi^2$  = 13.6111, df = 1; pH 2.5— $\chi^2$  = 5.5430, df = 1) and 30 minutes (pH 1.5— $\chi^2$  = 15.5556, df = 1; pH 2.0— $\chi^2$  = 12.0000, df

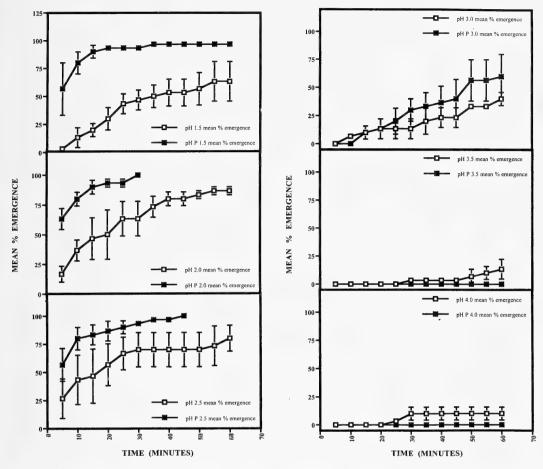


Figure 1. Effect of pH and pepsin on mean % distome emergence from the tail of *Proterometra macrostoma* over 60 minutes at 20°C. Means represent the average % emergence ± standard error (SE) from the three replicates with 10 cercariae/replicate. (pH = acidified saline only; pH P = acidified saline + 0.5% pepsin)

= 1; pH 2.5— $\chi^2$  = 4.320, df = 1). Emergence of *P. macrostoma* appeared to be greatly enhanced by the addition of pepsin to the most acidic pH's (i.e., 1.5–2.5). No significant differences were noted between these two treatments at the less acidic pH's (i.e., 3.0, 3.5, and 4.0), and overall emergence was minimal (Figure 1).

Within 15 minutes PI, all distomes had emerged from their cercarial tails in experimental infections of bluegill. No obvious trends for subsequent size increases were noted for worms in such infections other than changes in maximum egg size (Figure 2). The mean number of eggs/worm also showed a gradual increase from day 0 (13.8) to days 14—

18 (120.7) PI, followed by a slight decline in number between days 21–24 PI (Figure 3).

Changes in the general morphology/development of eggs were noted over the course of the experiment. Stage I eggs were the only eggs observed in adult worms from days 0 to 9 PI. After day 9 PI, a transformation into Stage II eggs was observed. It should be noted that Stage I egg production continued during this second stage, and thus two "populations" of eggs were present in worms at this time. Stage III (mature) eggs were apparent by day 18 PI. All three egg stages were observed in worms by this time; mature eggs were dispersed at the anterior end of the adult worm; immature eggs, at the posterior end.

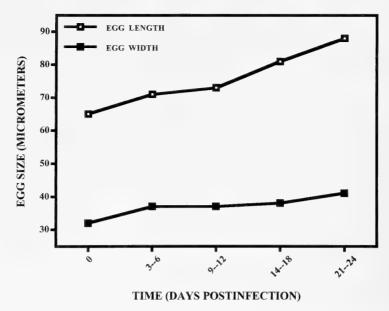


Figure 2. Mean lengths and widths of largest eggs in developing distomes of  $Proterometra\ macrostoma$  from experimental infections of bluegill,  $Lepomis\ macrochirus$ , over 24 days postinfection at 24.6°C. (number of eggs measured: day 0, n = 17; days 3–6, n = 42; days 9–12, n = 30; days 14–18, n = 33; days 21–24, n = 30)

#### DISCUSSION

Trematodes in the family Azygiidae are unusual in that they bypass a metacercarial stage, which is normally encysted in host tissue. Worms in this family are "encysted" in the tail of their own cercaria; thus there is no host tissue to be digested. Only triggers in the stomach (e.g., pH and pepsin) seem essential for emergence and subsequent establishment of the distome.

The high percentage of distome emergence in more acidic salines (pH 1.5-2.5) with 0.5% pepsin at 5 minutes and at 30 minutes (Figure 1) was similar to the rapid emergence observed in the experimental infections of bluegill at day 0 PI. In vivo, some P. macrostoma were recovered as emerged distomes at 5 minutes PI, and all had emerged from cercarial tails by 15 minutes PI. The similarity in rate of emergence in vivo vs. in vitro suggests that a pH range of 1.5-2.5 with a pepsin concentration of 0.5% approximates the conditions inducing emergence of the worm in the fish stomach. This is further corroborated by the work of Norris et al. (1973), who examined the rates of gastric acid and pepsin secretion into the gastric juice of bluegills. They found that bluegill gastric juice decreased from a pH of 5 to a pH of 1–2 immediately following ingestion of a simulated meal. Pepsin activity would be most pronounced in such a strongly acidic environment, further enhancing distome release from the cercarial tail.

Cercariae of *P. dickermani* (Anderson and Anderson 1963) and *P. autraini* (LaBeau and Peters 1995) contain eggs with fully developed miracidia, while only undeveloped eggs have been reported in cercariae from other species in this genus (Anderson and Anderson 1967; Horsfall 1934; Smith 1936). It has been suggested that a signal from the fish definitive host triggers maturation in some of these latter species of *Proterometra* (Braham et al. 1996). This "signal" must be focused on increased egg production as well as on egg maturation; other structures within these distomes appear to be fully developed following their emergence from the cercarial tail.

Dickerman (1934) observed that the only differences between the distome of the *P. macrostoma* cercaria and that of the adult worm were size and number of eggs present in utero. Similarly, Horsfall (1934) noted that, "the numerous eggs of the adult (*P. macrostoma*) mask certain structures which are conspicuous in the larval distome; otherwise the

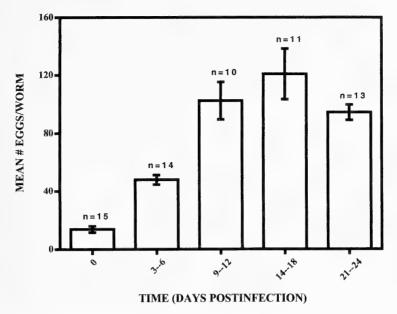


Figure 3. Mean  $\pm$  SE eggs/Proterometra macrostoma distomes obtained from experimental infections of bluegill, Lepomis macrochirus, over 24 days postinfection at 24.6°C. (n = number of worms assessed at each time interval)

general appearance (of the worm) is the same." Our observation of increased egg production in *P. macrostoma* over the initial 2.5 weeks of infection in bluegill corroborates these observations. Increased egg production also occurs in *P. albacauda* (Anderson and Anderson 1967), *P. catenaria* (Anderson and Anderson 1967), *P. edneyi* (Uglem and Aliff 1984), *P. sagittaria* (Dickerman 1946), and *P. septimae* (Anderson and Anderson 1967) following release from their cercarial tails in the definitive host.

Egg production appears to be continuous once the distome of *P. macrostoma* is released in the fish stomach as evidenced by (1) the steady increase in egg number over time (up to day 18 PI) and (2) the presence of Type I eggs (i.e., early cleavage) in older fish infections. Decrease in egg number between days 21–24 PI may coincide with initiation of egg release from the adult worm into the host digestive tract, but further work will be required to verify this.

In our study, *P. macrostoma* eggs required 18–24 days at 24.6°C to develop miracidia. Horsfall (1934) indicated that 15–30 days are required depending on the number of eggs contained within the digenean prior to its emergence from the cercarial tail. In addition

to the progenetic state of these distomes, proposed *P. macrostoma* strain differences (Dickerman 1945; Riley and Uglem 1995) may affect maturation time.

Current work is being conducted in our lab using molecular techniques to determine the validity of these *P. macrostoma* strains.

## ACKNOWLEDGMENTS

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# New State Records and New Available Names for Species of Kentucky Moths (Insecta: Lepidoptera)

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#### **ABSTRACT**

The authors add records of 35 moth species to the list of Lepidoptera known in Kentucky, bringing the total to 2423 from the 2388 published in the Covell (1999) annotated checklist. These are in the families Gracillariidae (1), Oecophoridae (1), Gelechiidae (3), Tortricidae (4), Crambidae (16), Pterophoridae (2), Geometridae (1) and Noctuidae (7). In addition scientific names are given for the three "Chionodes undescribed species" (Gelechiidae) listed as such by Covell (1999) and recently made available by Hodges (1999).

#### INTRODUCTION

Intensive collecting, identification, and monitoring of the moth and butterfly fauna of Kentucky culminated in December 1999 with the publication of Covell (1999), in which 2388 species of moths and butterflies were documented. Even at that time there were additional species to add. And since that publication came out names have been made available for three species that were indicated under generic headings as "species" because we could not cite unpublished names. Thirtyeight species are added to the state list in this paper, bringing the total to 2426. This is the first supplement to Covell (1999).

We thank the following specialists for identifying specimens from which most of these records resulted: Bernard Landry, William E. Miller, Herb Neunzig, Eric L. Quinter, Michael Sabourin, David Wagner, and Reed Watkins.

Below are listed the new additions and names in order of the numbers assigned in the Hodges et al. (1983) checklist. Specimens on which these identifications are based are in the Lovell Insect Museum at the University of Louisville and in the private collections of those cited as the collectors. Those cited from Hodges (1999) are located in the National

Museum of Natural History, Washington, D.C., and the Philadelphia Academy of Natural Sciences collection.

#### GRACILLARIIDAE

0606 Caloptilia fraxinella (Ely)

Boone Co., Big Bone Lick State Park, larva collected on *Fraxinus 4* Jun 1995; adult emerged 5 Jun 1995; D.J. Wright.

#### **OECOPHORIDAE**

1015 Antaeotricha osseela (Wlsm.)

Laurel Co., junction of forest service roads 121 and 4158, larva collected on *Quercus montana*, 18 May 1996, adult emerged 31 Jul 1996; Rowan Co., east end of Clack Mountain Road West, 26 Aug 1994; both collected by D.J. Wright.

## **GELECHIIDAE**

2061.1 Chionodes hapsus Hodges

A paratype female of this species was listed by Hodges (1999, p. 56) from Fleming Co., Fleming, 31 May 1938, A.F. Braun. This is not one of the "C. undescribed species" listed in Covell (1999, p. 37).

2061.3 Chionodes suasor Hodges

A paratype male of this species was listed by Hodges (1999, p. 58) from Rowan Co., Morehead, 30 Jun 1960, collected by Lewis and Freeman. This is not one of the "C. undescribed species" listed in Covell (1999, p. 37).

2119.2 Chionodes sevir Hodges

This name validates the entry under this checklist number in Covell (1999, p. 37). A paratype male is listed in Hodges (1999, p. 138) with the following data: Fulton Co., Fulton, 19 Sep 1975, C. C. Cornett. See Covell (1999, p. 37).

2119.1 Chionodes baro Hodges

A paratype male is listed by Hodges (1999, p. 145) from Bell Co., Pine Mountain State Park, 18 Jul 1975, A.J. Brownell. This is the name that validates the entry under this checklist number in Covell (1999, p. 37). The designation "Harlan Co." in that entry was an error.

2119.3 Chionodes adamas Hodges

This is the name that validates the entry under this checklist number in Covell (1999, p. 37). Paratype data as published by Hodges (1999, p. 151) follow: "Clack Mountain, Rowan County; 19, 22 June 1941; A.F. Braun (2 females). Otter Creek Park, Meade County, 8 October, 1979, C.V. Covell Jr. (1 female). Same locality, 17 March, 1987, B.S. Nichols (1 male). Rd 9B, Indian East Fork, Kelley Br., 720', Manifee [sic] County; 9–18 August, 1985; J.S. Nordin (2 males, 1 female). Tunnel Ridge Road, Powell County; 4–11 March, 1989; D.J. Wright (1 male, 2 females)."

2066.1 Chionodes aruns Hodges

Hodges (1999, p. 189) described this species from Texas, and although he lists Kentucky specimens of the species, they were not included in the paratype series. Their data are quoted as follows: "Cumberland National Forest, Pulaski County, Kentucky; 26 April 1939; A.F. Braun (1 female). Lick Fork, Rowan County, Kentucky; 4 April 1938; A.F. Braun (1 male). Cascade Caves, Carter County, Kentucky; 5 May 1956; A.F. Braun (1 female)."

# TORTRICIDAE

2717 Endopiza yaracana (Kft.)

Powell Co., Red River Gorge, Tunnel Ridge Road; 3 May 1991, D.J. Wright.

2795 Olethreutes tiliana (Heinr.)

Boone Co., Middle Creek Road, larvae collected on *Tilia neglecta* 5 May 1993, two females emerged on 30 May 1993; Laurel Co., Rockcastle Campground, larva collected on *Tilia* 1 May 1993, adults emerged 27 May and 3 and 10 Jun 1993, D.J. Wright; Laurel Co., Daniel Boone National Forest, Rockcastle Recreation Area, larva on *Tilia neglecta* collected 1 May 1993, two females emerged 25 and 28 May 1993.

2885 Rhyacionia aktita W.E. Miller
Laurel Co., Daniel Boone National Forest, Forest
Service Road 131, 2 miles from State Road 3497,
19 Apr 1992, D.J. Wright; powerline corridor east
side of south end of Forest Service Road 775, 11

Apr 1997, L.D. Gibson; Forest Service Road 615a, 30 Apr and 4 May 1996, D.J. Wright.

3550 Acleris youngana (McDunnough) Harlan Co., summit of Big Black Mountain, 12 Jul 1980, L.D. Gibson.

#### CRAMBIDAE

4754 Synclita tinealis Munroe

Henderson Co., Frank Sauerheber Unit, Sloughs Wildlife Management Area, 22 Aug 1992, L.D. Gibson.

4769 Neargyractis slossonalis (Dyar)

plant), L.D. Gibson.

Hopkins Co., 2 miles SE Dawson Springs along Caney Creek, 20 Aug 1999 in light trap, L.D. Gibson. This record is a significant northern range extension. Munroe (1972, p. 116) had records only from Florida but believed it would be recorded in neighboring states.

4981 Helvibotys pseudohelvialis (Capps)
 Fulton Co., Willingham Bottoms, Rt. 94, 2.5
 miles E of Cayce, 8 Sep 1991.

5034 Pyrausta signatalis (Wlk.)
 Laurel Co., Daniel Boone National Forest, powerline corridor east side of south end of Forest Service Road 775, 30 May, 27 Jun, and 10 Jul 1997 at UV and MV lights near Monarda (host

5173 Diasemiodes nigralis (Fernald) Bath Co., Cave Run Recreation Area, Forest Service Road 918, 5 Sep 1987, D.J. Wright.

5248 Lygropia tripunctata (Fabricius) Henderson Co., Frank Sauerheber Unit, Sloughs National Wildlife Area, 10 Sep 1983, C.V. Covell Jr.

5450 Parapediasia decorella (Zinck.) Powell Co., Natural Bridge State Park, 7 Jul 1981, in light trap, C.C. Cornett.

5653 Acrobasis vaccinii Riley Laurel Co., junction of Forest Service roads 121 and 4158; 18 May 1996; Forest Service Road 131, 30 May 1992; D.J. Wright.

5766 Immyria nigrovittella Dyar
 Laurel Co., Daniel Boone National Forest, Forest
 Service Road 615a, 22 Apr 1995 and 4 May 1996,
 D.J. Wright; powerline corridor E side of S end
 of State Road 775, 30 May 1997, L.D. Gibson.

5775 Salebriaria tenebrosella (Hulst)
Rowan Co., County Road 1274, 2 miles W of Rt.
519, 16 Jul 1994, L.D. Gibson; Rowan Co., 3.3
miles S of Rt. 519, and also east end of Clack
Mtn. Rd. West, 26 Aug 1994, D.J. Wright.

5777.1 Salebriaria atratella Blanchard & Knudson Laurel Co., Bolton Branch, 18 May 1996, D.J. Wright.

Nephopterix vetustella (Dyar)
 Boone Co., Big Bone Lick State Park, 5 May
 1980; Powell Co., Tunnel Ridge, Red River Gorge

Geological Area, 19 Jun 1993; both collected by L.D. Gibson.

Nephopterix crassifasciella Ragonot
 Laurel Co., Daniel Boone National Forest, powerline corridor, E side of S end of State Road 775,
 Jun and 10 Jul 1997, L.D. Gibson.

5944 Homoeosoma deceptorium Heinrich Boone Co., Camp Ernst, 17 Aug 1979, L.D. Gibson.

5953 Laetilia fiskeella Dyar
 Laurel Co., Daniel Boone National Forest, powerline corridor, E side of S end of State Road 775,
 17 May and 27 Jun 1997 at lights, L.D. Gibson.

6122 Stenoptilodes brevipennis (Zeller) Bullitt Co., Bernheim Arboretum and Research Forest, 20–24 Apr 1976 in Malaise trap, A.J. Brownell.

#### PTEROPHORIDAE

6107 Gillmeria pallidactyla (Haworth) Harlan Co., summit of Big Black Mountain, 28 Jun 1999, male and female, Reed A. Watkins.

Oidaematophorus eupatorii (Fernald)
 Harlan Co., Big Black Mountain, 14 Jul 1979;
 Pine Mountain Settlement School, 4 Jul 1977;
 both collected at black light by C.V. Covell Jr.

#### GEOMETRIDAE

6851 Philtraea monillata Buckett
Carlisle Co., Sandy Branch, Burkley, 4 Sep 1999,
numerous specimens in light traps, C.V. Covell
Jr., L.D. Gibson, and others.

#### NOCTUIDAE

8658 Selenisa sueroides (Guenée) Carlisle Co., Sandy Branch, 10 Oct 1999, in light trap, William R. Black Jr.

9386 Luperina trigona Smith
Larvae were collected from Arundinaria (cane)
stalks by W.R. Black Jr. and Eric Quinter 10–18
May 1999 in the following localities: Ballard Co.,
Stovall Creek; Carlisle Co., Sandy Branch near
Burkley; Fulton Co., Willingham Bottoms, Rt. 94,
2.5 miles E of Cayce and Reelfoot Lake National
Wildlife Area; Graves Co., Boaz; Livingston Co.,
Burna; McCracken Co., Massac Creek Bottoms,
Paducah. An adult was collected in a light trap in

McCracken Co., 1 mile W of Clinton Rd., 4 Sep 1999 by W.R. Black Jr.

9329.1 Apamea undescribed species K. Mikkola Bullitt Co., Bernheim Arboretum and Research Forest, 12–18 Jul 1976 in Malaise trap, A.J. Brownell; Jefferson Co., Valley Station, "Aug.", S. Scholz; Russell Co., Lake Cumberland State Park, 11 Jun 1980, C.C. Cornett.

9419 Oligia mactata (Guenée) Jefferson Co., Valley Station, 26, 29 Sep, 2–5 and 20–29 Oct 1997, S. Scholz.

9425 Meropleon cosmion Dyar
 McCracken Co., Paducah, Massac Creek Bottoms
 in cane, 13 Nov 1999 in light trap, W.R. Black Jr.
 9681.1 Elaphria cornutinis Saluke & Pogue.

Two male paratypes were listed from Kentucky by Saluke and Pogue (2000): Metcalfe Co., Highway 218 north of Center, 25 Apr and 6 May 1994, C. Cook. This species is very similar to *E. festivoides* (Gn.). Some records listed under *E. festivoides* in Covell (1999, p. 160) will turn out to be this newly described species.

10460 Leucania calidior (Forbes)
Carlisle Co., Sandy Branch near Burkley, 2 Oct
1999, in light trap, W.R. Black Jr.; Livingston Co.,
Burna, 10–18 May 1999, larva in cane reared to
adult, E.L. Quinter and W.R. Black Jr. The species name is misspelled as "callidior" in Hodges
et al. (1983) (fide E.L. Quinter).

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# Comparative Effects of Zinc, Lead, and Cadmium on Body and Tissue Weights of Weanling, Adult, and Aged Rats

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#### ABSTRACT

The effects of feeding various levels of Zn in the diet and Pb and Cd in drinking water were determined in weanling, adult, and aged rats for 4 or 8 weeks. Zn levels in the diet were: Zn-deficient diet or Zn0; 60 mg/kg high Zn diet (Zn60) for experiment 1 & 3; 4 mg/kg low Zn diet (Zn4); 24 mg/kg high Zn diet (Zn24) for experiment 2; and 12 mg/kg Zn diet as control. Zn0 diets reduced feed intake (FI) and body weight (BW) in weanling rats by 35% and 80% and BW in aged rats by 60%; BW of Zn4 adult rats decreased by 40% ( $P \le 0.05$ ). Feed intake of adult and aged rats was comparable among all groups. Feed efficiency (FE) decreased fourfold and twofold in weanling and adult rats, respectively. Liver and kidney weights were significantly lower in weanling group fed Zn0 diet as compared to the control or Zn60 groups ( $P \le 0.05$ ). Weanling and adult rats given 20 mg/liter Pb and 5 mg/liter Cd in drinking water had lower water intake (WI) than the control ( $P \le 0.05$ ). These results indicate that Zn0 diet decreased FI, BW, and FE in weanling rats; Zn0 and Zn4 diets reduced BW in adult and aged rats but without a decline in FI. Lead and Cd in drinking water did not affect growth or FI but decreased WI in weanling and adult rats. Although lead and cadmium did not modify growth in either zinc-deficient or low zinc-fed rats, these diets decreased growth and feed efficiency, which may partly be attributed to loss of appetite and altered Zn homeostasis.

#### INTRODUCTION

Trace elements have an important and critical role in maintaining nutrition and normal health in animals. Zinc is essential for reproduction, growth, and development. As a component of over 200 enzymes, it plays a vital role in cell replication and differentiation and in the structure and function of cellular membranes. The metabolic and physiological status of zinc could be modified or exacerbated by the ingestion of toxic metals such as lead or cadmium, which are widespread contaminants of the food chain and potable water sources (Underwood 1979). The cumulative retention of lead and cadmium after chronic low-level exposure can result in manifestations of toxicity such as disturbance of heme synthesis, dysfunction of the nervous system, and renal damage (Friberg et al. 1985). Concurrent exposure to lead and cadmium may increase toxicity of one or both metals and produce significant changes in the metabolism of zinc (Mahaffey et al. 1982).

Coppen-Jaeger and Wilhelm (1989) reported inhibition of zinc absorption in isolated rat intestinal preparations after low-level cadmium exposure. While high levels of zinc may

reduce some of the toxic effects of cadmium (Sato and Nagai 1989), zinc deficiency may enhance toxic effects of even low levels of cadmium (Kunifuji et al. 1987; Tanaka et al. 1995). Zinc is reported to have a protective effect on lead toxicity when both are present in the diet at high levels due to inhibition of lead absorption (Cerklewski and Forbes 1975). Age may affect zinc metabolism because related changes in body composition are often paralleled by a decline in physiological and metabolic functions, which could also modify growth and food conversion efficiency (Sandstead et al. 1982). Age-related differences in response of rats to lead or cadmium exposure were reported by Cory-Slechta et al. (1989) and Song et al. (1986). However, most published research has been done with pharmacological or toxic doses of lead or cadmium using either oral (Sato and Nagai 1989) or parenteral methods (Kunifuji et al. 1987) of exposure, which have little bearing on exposure conditions in the environment and may not be of much practical significance (Sabbioni et al. 1978). Therefore, we hypothesize that low-level exposure to lead and cadmium would not alter zinc absorption and metabolism. To test this hypothesis, we have determined the interaction among lead, cadmium, and zinc in

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growing, adult, and aged rats fed increasing levels of zinc in the diet.

In this paper, we present data on the interaction of low-level lead and cadmium in drinking water on growth, feed intake, feed efficiency, and tissue weights of weanling, adult, and aged rats fed different levels of zinc in the diet

#### MATERIALS AND METHODS

Male Sprague Dawley rats (Harlan Sprague Dawley, Inc.) 27 days, 8 weeks, or 18 months old were housed singly in suspended polypropylene cages on stainless steel racks in temperature controlled rooms with 12 h light/dark cycles. During a 1-week period of acclimation, rats were fed the control diet and given distilled drinking water. The experimental diets (Harlan Teklad, Madison, WI) containing different levels of zinc were based on modifications of the zinc-deficient diet (AIN93 formula), using egg white as the protein source and a mixture of starch and dextrin as the carbohydrate source (Reeves et al. 1993). Zinc sulfate was added to experimental diets as follows: Control group = 12 mg/kg (Zn12); zincdeficient group = 0 mg/kg (Zn0); low-zinc group = 4 mg/kg (Zn4); and high-zinc group = 24 mg/kg (Zn24) or 60 mg/kg (Zn60). Alldiets were analyzed for their zinc content by atomic absorption spectrophotometry, which was within a 5% range of the expected level. Zinc content of the diets as mg Zn/kg diet were Zn0 = 0.8, Zn4 = 4.9, Zn12 = 12.7, Zn24 = 25.0, and Zn60 = 60.5.

After the acclimation period, the rats were randomized and grouped such that the average initial body weights were similar in all groups (six rats/group; the deficient and pairfed groups had weight-matched pairs) and were assigned to the experimental diets described above. The rats were given distilled water containing 10 mg/liter sodium as NaCl, or 20 mg/liter Pb as lead acetate, or 5 mg/liter Cd as cadmium chloride (0 = control). Rats had free access to feed and water as follows: experiment 1 and 3, 30 days and experiment 2, 60 days. In experiment 1 (72 wearling) and experiment 3 (36 aged) rats were fed diets Zn0, Zn12, Zn60, or Zn12PF (pair-fed control diet daily to the intake of the Zn0 group); in experiment 2 (72 adult) rats were fed diets Zn4, Zn12, Zn24, or Zn12PF (pair-fed control diet daily to the intake of the Zn4 group). Feed intake (FI) was determined daily, whereas body weight (BW) and water intake (WI) were recorded weekly. Feed efficiency (FE) was calculated as the ratio of FI to weight gain (WG). The three experiments were conducted separately under uniform conditions. For ease of handling 72 rats, experiments 1 and 2 were split in half such that each group had 3 rats, and each batch was run under identical conditions, 1 week apart. The procedures for these animal experiments were approved by the Kentucky State University Animal and Human Welfare Committee.

At the end of the experimental period, rats were terminated under diethyl ether anesthesia for removal of liver and kidneys. The organs were cleansed of extraneous tissue, weighed, and stored at  $-70^{\circ}$ C. All data were statistically analyzed by ANOVA using the SAS program and a two-way, 4 (Zn levels)  $\times$  3 (Na/Pb/Cd levels) factorial design. Significant differences between means were obtained by the Duncan's multiple range test.

#### RESULTS

The general appearance of the animals was influenced by dietary zinc. Weanling rats were especially affected by zinc deficiency as they lost abdominal hair and had hairless patches elsewhere on the body; those fed Zn12 and Zn60 had smooth hair. Adult rats fed low zinc diet (Zn4) did not lose hair but the texture of the hair was rough. No physical difference in appearance was noted in aged rats. Growth, FI, FE, and tissue weight data are presented here.

Since exposure to oral lead and cadmium did not influence BW or FI, all data presented in Figure 1 were merged ignoring lead and cadmium exposure (n = 18). Weanling rats fed Zn0 and Zn12PF diets had significantly lower FI (Figure 1A), weight gain (WG) (Figure 1B), and FE as compared to control (Figure 1C;  $P \le 0.05$ ): Feed intake was about 35% lower, while WG was reduced by 80% and 54%, respectively. Weanling rats fed diet Zn60 grew throughout the 4-week experiment and their Fl, WG, and FE were comparable to those of the control group. Feed efficiency was lower among rats fed Zn0 and Zn12PF, and both were two-fold and four-fold lower than the control. Growth and Fl data of adult

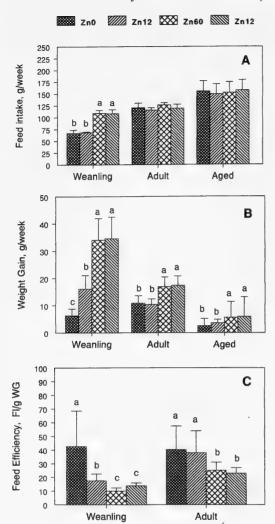


Figure 1. Feed intake (A), weight gain (B), and feed efficiency (C) of weanling, adult, and aged rats fed Zn0 (Zn4 for adult rats), Zn12PF, Zn60, and Zn12 (control) for 4 weeks (8 weeks for adult rats). Means  $\pm$  SD; n = 18 (weanling and adult groups). Means  $\pm$  SD; n = 9 (aged groups). Means in different diet groups not sharing the same superscript are significantly different at  $(P \le 0.05)$ .

rats fed low and high zinc diets for 8 weeks are presented in Figure 1A, 1B, 1C. Weight gain and FE of rats fed Zn4 and Zn12PF diets were 40% and two-fold lower than those fed the control diet ( $P \le 0.05$ ), respectively. There was no significant difference in Fl among all the groups ( $P \le 0.05$ ). Aged rats showed a similar BW and Fl pattern as adult rats (Figure 1A, 1B, 1C). Feed intake was comparable

among aged rats in the different diet groups irrespective of zinc level, but WG was 60% and 43% lower in Zn0 and Zn12PF groups than the control or Zn60 group ( $P \leq 0.05$ ). Rats (especially the Zn0 fed groups) experienced a day-to-day increase or decrease in feed consumption, which indicated cycling of feed intake. Due to wide variation in Fl, FE values of aged rats could not be calculated accurately and therefore are not reported here. Low-level oral exposure to lead or cadmium had no effect on growth and Fl of weanling, adult, or aged rats.

Tissue weights of rats are presented in Table 1. In weanling rats (experiment 1), average liver weights were about 54% lower in the Zn0 and Zn12PF groups than in the control or Zn60 groups ( $P \le 0.05$ ). Similarly, kidney weights were a third lower in the Zn0 and Zn12PF groups compared to the control ( $P \le 0.05$ ). Also, mean liver and kidney wet weight/body weight ratios were comparable among all dietary groups. Such differences in tissue weights were not observed in adult or aged rats. Lead and cadmium exposure had no effect on tissue weights.

Water intake data of rats for experiments 1 and 2 are presented in Table 2. There was significant interaction between dietary zinc levels and the concentration of lead and cadmium in drinking water, especially in the zinc-deficient group. Water intake was significantly lower in weanling and adult rats given lead and cadmium in drinking water than those given sodium in water (control), regardless of the level of zinc in the diet  $(P \le 0.05)$ . The data also indicated that, in weanling rats, WI was significantly influenced not only by the presence of lead and cadmium in drinking water but also by the amount of zinc in the diet. Water intake for experiment 3 could not be accurately measured because of excessive spillage, so these data are not presented.

#### DISCUSSION

Cellular homeostasis, a major regulator of zinc metabolism, ensures that over a wide range of zinc intake, tissue or cellular zinc levels are maintained at physiological concentrations by either enhancing absorption or decreasing loss through the gastrointestinal tract (Golden 1989). In our study, we found that Zn0 diet significantly decreased feed intake,

Table 1. Tissue weights of wearling (n = 18), and aged (n = 9) rats fed zinc deficient (Zn0), high zinc (Zn60), or control (Zn12) zinc diets for 4 weeks, and adult rats (n = 18) fed low zinc (Zn4), high zinc (Zn24), and control zinc (Zn12) for 8 weeks. Data are presented as Mean ± SD.

			Zinc levels in the diet						
Parameters	Experiment	Zn 0	Zn12PF¹	Zn 60	Zn12'				
Liver, g	1 (W) <sup>2</sup>	$5.4 \pm 0.8^{\rm b}$	$5.1 \pm 1.1^{b}$	$9.5 \pm 1.5^{a}$	$9.7 \pm 1.3^{a}$				
	2 (A)	$11.2 \pm 1.5$	$10.4 \pm 1.1$	$11.6 \pm 1.5$	$11.8 \pm 1.5$				
	3 (AG)	$17.8 \pm 2.3$	$16.8 \pm 2.4$	$17.9 \pm 0.8$	$17.2 \pm 2.5$				
Kidney, g	$1 (W)^2$	$1.2 \pm 0.1^{\rm b}$	$1.3 \pm 0.2^{\rm b}$	$1.9 \pm 0.2^{a}$	$1.9 \pm 0.2^{a}$				
, 0	2 (A)	$2.2 \pm 0.1$	$2.1 \pm 0.2$	$2.2 \pm 0.02$	$2.2 \pm 0.2$				
	3 (AG)	$3.9 \pm 0.9$	$3.1 \pm 0.5$	$3.2 \pm 0.01$	$3.2 \pm 0.4$				
Liver wt/body wt ratio	$1 (W)^2$	$0.04 \pm 0.007$	$0.03 \pm 0.005$	$0.04 \pm 0.002$	$0.04 \pm 0.003$				
,	2 (A)	$0.03 \pm 0.004$	$0.03 \pm 0.002$	$0.03 \pm 0.002$	$0.03 \pm 0.003$				
	3 (AG)	$0.04 \pm 0.005$	$0.03 \pm 0.005$	$0.04 \pm 0.003$	$0.03 \pm 0.004$				
Kidney wt/body wt ratio	$1 (W)^2$	$0.01 \pm 0.001$	$0.01 \pm 0.001$	$0.01 \pm 0.001$	$0.01 \pm 0.001$				
,	2 (A)	$0.01 \pm 0.001$	$0.01 \pm 0.005$	$0.01 \pm 0.004$	$0.01 \pm 0.003$				
	3 (AG)	$0.01 \pm 0.003$	$0.01 \pm 0.001$	$0.01 \pm 0.001$	$0.01 \pm 0.001$				

Data were grouped ignoring Pb, Cd, exposure as no significant differences were observed between these groups. Means in rows with letter superscripts indicate significant differences; absence of letters indicates means are not significantly different at  $P \le 0.05$ .

Rats in pair-fed (Zn12PF) control group were fed control diet (Zn12) in the amount equal to that eaten by either Zn0 or Zn4 groups.

We weanling; A = Adult; AG = Aged rats (see materials and methods for details.

body weights, and feed conversion efficiency of weanling rats and that long-term (8 week) Zn4 feeding also reduced body weights without a concomitant decline in Fl, which suggests alteration of zinc homeostasis or a decrease in the availability of zinc for growth and maintenance. While exposure to low levels of lead and cadmium did not affect Fl or BW in any age group, it decreased WI significantly in weanling and adult rats.

Guigliano and Millward (1984) reported that in zinc deficient animals, zinc may be redistributed from bone and conserved in muscles, being released only for the growth and vital functioning of essential tissues in a catabolic state. It is not clear if the failure of zincdeprived rats to grow is a result of zinc on appetite or an effect of impaired cell division on growth (O'Dell and Reeves 1989). Rains and Shay (1994) initially suggested that reduced Fl in Zn deficient rats could be because of impaired function of neuropeptide Y, a known stimulator of appetite and carbohydrate consumption. More recently, neuropeptide Y has been shown to increase in zinc deficiency, probably to restore normal FI, but this is prevented by receptor binding of this peptide (Lee et al. 1998). Our data suggest that reduction in growth could be due to reduced Fl as well as impaired cell division chiefly in weanling rat, since pair-fed groups lost less weight than did the Zn0 (weanling) and

Table 2. Water intake of weaning rats fed zinc-deficient (Zn0) or high zinc (Zn60) diets for 4 weeks, or adult rats fed low zinc (Zn4) or high zinc (Zn24) diets for 8 weeks, and exposed to lead or cadmium in drinking water. Means ± SD; n = 18/group.

		Zinc levels in the diet (ml water intake/wk						
Experiment	Metals in water	Zn 0	Zn 12 PF1	Zn 60	Zn121			
1 (W) <sup>2</sup>	Na	130 ± 7a,f	140 ± 28a,ef	161 ± 13a,e	$178 \pm 16^{a.c}$			
, ,	Pb	$100 \pm 22^{\rm b,f}$	$131 \pm 10^{\rm b,e}$	$153 \pm 9b,d$	$162 \pm 15^{b}$			
	Cd	$89 \pm 20^{c,f}$	$118 \pm 16c,e$	$150 \pm 13b,d$	$152 \pm 14^{b}$			
$2 (A)^2$	Na	$180 \pm 27^{4}$	$167 \pm 35^{a}$	$160 \pm 15^{a}$	$166 \pm 20^{a}$			
	Pb	$154 \pm 20^{b}$	$149 \pm 20^{\rm b}$	$160 \pm 11^{a}$	$148 \pm 11^{a}$			
	Cd	$150 \pm 25^{\text{b}}$	$144 \pm 20^{\rm b}$	$130 \pm 15^{b}$	$144 \pm 13^{b}$			

ANOVA indicates interaction between zinc levels, and lead and cadmium in water. a, b, c, denote significant differences in each row; d, e, f, denote significant differences in each column (P \u2224 0.05), within each experiment

Rats in pair-fed (Zn12PF) control group were fed control diet (Zn12) in the amount equal to the intake by either Zn0 or Zn4 groups. (See materials and methods for details).

<sup>&</sup>lt;sup>2</sup> W = weanling; A = Adult. Data for Experiment 3 are not available.

Zn4 (adult) rats, respectively. Thus, the higher weight loss in the Zn0 group may be attributed partly to zinc deprivation and lowered Fl; that in the pair-fed group, to lowered Fl.

Numerous reports (Abdel-Mageed and Oehme 1991; Faraji and Swendseid 1983; Guigliano and Millward 1984) attest to the significant decline of Fl and growth in Zn0 weanling rats and support our observations. In our study, aged rats on Zn0 diet for 4 weeks also lost weight but, unlike weanling and adult rats, without a significant decrease in Fl. One reason for this may be the variation and cycling of Fl we found in all groups of aged rats irrespective of zinc level in the diet; whereas, cycling of FI was found only in zinc-deficient weanling and adult rats. However, recent studies analyzing food-intake patterns of zinc deficient rats have shown that the characteristic cyclical variation in FI and body weight changes were found not only by group but also in each individual rat fed zinc-deficient diets (Tamaki et al. 1995) and that body weight change is generally well synchronized with that of FI (Aiba et al. 1997). The reduction in feed conversion efficiency we found in weanling rats was reported previously by William and Mills (1970). Conflicting reports in the literature suggest that zinc levels as high as 1000 µg/g diet fed for 8 weeks do not alter Fl or growth, while zinc levels up to 2500 µg/g diet for 3 weeks decrease growth (Abdel-Mageed and Oehme 1991; Song et al. 1986; Story and Greger 1987). Toxicity of such high zinc levels may have influenced their results. Our results support those of Abdel-Mageed and Oehme (1991), since moderately high zinc (60 µg/g) diets did not produce any change in growth or

In our study, low levels of lead and cadmium in drinking water did not alter feed intake or body weight when dietary zinc levels were deficient or low or when the diets contained high zinc levels, indicating a lack of interaction between zinc and toxic metals. Previous reports have shown that high levels of lead and cadmium administered through drinking water or parenterally reduce zinc absorption and thereby body weight (Kunifuji et al. 1987; Tanaka et al. 1995) or increase the toxic effect of cadmium (Sato and Nagai 1989). High zinc intake is known to protect against toxic effects of lead (Cerklewski and Forbes 1975). In rats fed normal zinc diets, Kirchgessner et al. (1987) found that age did not modify the effects of increasing levels of lead or cadmium in the diet. This report supports our observations that at low levels of lead or cadmium exposure there is no interaction between dietary zinc levels or age of the animal on body weight and feed intake.

Although organ weight is known to decline with BW, higher organ to BW ratios in weanling rats fed zinc-deficient diets have been reported (Abdel-Mageed and Oehme 1991; Meydani et al. 1983), but our data do not completely agree with these observations since we found that, in weanling rats, liver and kidney weights were significantly lower in Zn0 or pairfed groups as compared to the controls. This parallel and proportional decrease or increase in organ and body weights is indicated by the similar kidney and body weight ratios of all groups in the three experiments irrespective of age; it refutes the effect of zinc deficiency on decline of only skeletal mass suggested by Abdel-Mageed and Oehme (1991) who did not include a pair-fed group for comparison. Our observation that lead and cadmium had no significant effect on organ weights in all age groups is similar to reports by Rader et al. (1981), Vyskocil et al. (1991), and Cory-Slechta et al. (1989) in weanling, adult, and aged rats, respectively. Age-related changes begin to accelerate in rodents after 16 months of age (Cory-Slechta et al. 1989). The aged rats in our study were 18 months old and yet showed no adverse effects of short-term exposure to low lead and cadmium levels.

Addition of low levels of toxic metals in drinking water is assumed not to alter WI, but our data demonstrate that WI decreased significantly in both weanling and adult rats when low levels of lead and cadmium salts were added to drinking water. These data are largely supported by the reports of Vyskocil et al. (1990) in weanling rats, Kotsonis and Klaassen (1978) in adult rats, and Cory-Slechta (1990) in aged rats using much higher (10-100 times) concentrations of lead or cadmium than we used. We were unable to measure WI accurately in aged rats; thus the decrease in WI in this group cannot be confirmed. It is apparent that reduced FI produced by Zn0 diet also significantly decreased WI in weanling rats, while increasing Zn in the diet did not alter WI. We found no such changes in adult or aged rats, indicating a direct relationship between FI and WI in weanling rats.

From our data, it is apparent that zinc-deficient or low-zinc diets fed for a prolonged period can significantly decrease weight gain and feed conversion efficiency in weanling and adult rats, respectively. These effects may be attributed to impaired cell division and metabolism caused by altered Zn homeostasis due to Zn deficiency, as well as reduced Fl resulting from loss of appetite. Low-level oral lead and cadmium exposure was not toxic to all age groups of rats. Significant reduction in water intake after addition of lead or cadmium suggests that this may not be an appropriate method of oral exposure when quantitative toxic metal exposure is critical, as actual intake may vary significantly within and between groups. Addition of toxic metals to the diet may perhaps be a preferable method.

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# Rare and Extirpated Biota of Kentucky

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#### ABSTRACT

The Kentucky State Nature Preserves Commission has updated and revised the lists of rare and extinct or extirpated biota last published in 1996 and updated in 1997 and 1999. The newly revised lists include a lichen, and 389 plant and 282 animal taxa considered rare in Kentucky, and 4 plant and 42 animal taxa considered extirpated from Kentucky or extinct.

#### INTRODUCTION

The Kentucky State Nature Preserves Commission (KSNPC) is mandated to identify and protect natural areas to conserve Kentucky's natural heritage. To accomplish this mandate KSNPC works in cooperation with many scientific authorities in the public, private, and academic sectors. To help focus its conservation activities, KSNPC has developed a list of taxa native to the state that are considered rare. A list of species presumed extinct or extirpated from Kentucky is also maintained to document the loss of biodiversity, much of which is attributable to human activities. The overall goal of publishing these lists is to assist in the recovery and preservation of Kentucky's rich natural diversity.

KSNPC uses The Nature Conservancy's standardized Natural Heritage Program (NHP) methodology (TNC 1988) to manage distributional and ecological information on rare taxa, high quality natural communities, and other unique natural features in map, manual, and computer files. This information is used to locate aggregations of these entities for monitoring and protection. The NHP methodology is well suited for the revision process outlined below.

#### **METHODS**

Each taxon listed by KSNPC (1996, 1997, 1999), as well as other unlisted organisms, were evaluated to assign a conservation status. The evaluation criteria used included the number, age, and accuracy of occurrences; historical and present geographic distribution; habitat requirements; threats to the taxon including habitat loss; and ecological fragility. The information used to make the evaluation was that available as of 1 Jan 2000. The re-

sultant list and proposed status designations were submitted to knowledgeable individuals for peer review and suggestions for taxa to add and delete. All comments received were considered and in many cases discussed with the reviewer before the list was finalized.

Sources consulted for the plant and lichen names are: Anderson (1990); Crum et al. (1990); Egan (1987); and Kartesz (1994). The sources consulted for the common and scientific names of animals are as follows: gastropods-Hubricht (1985) and Turgeon et al. (1998); freshwater mussels—Gordon (1995) and Turgeon et al. (1998); crustaceans-Barr (1968), Holsinger (1972), Taylor and Sabaj (1998), USFWS (1994), and Williams et al. (1989); insects—Arnett (1983); Barr (1996), Cassie et al. (1995), Krekeler (1973), Mc-Cafferty (1996), Miller (1992), Morse (1993), Paulson and Dunkle (1999), Schuster (1997), and Schweitzer (1989); fishes—Ceas and Page (1997), Page and Burr (1991), Robins et al. (1991), USFWS (2000), and Warren (1992); amphibians and reptiles—Collins (1990), Frost (1985), and King and Burke (1989); breeding birds—AOU (1998); mammals— Hall (1981), Jones et al. (1992), and Wilson and Reeder (1993).

## Status Designations

The intent of assigning status designations is to (1) indicate the degree of rarity of the taxon, (2) indicate the degree of threat to the continued survival of the taxon, and (3) aid in establishing conservation priorities. The five KSNPC status designations defined below have no legal or statutory implication.

Endangered (E). A taxon in danger of extiration and/or extinction throughout all or a significant part of its range in Kentucky. Threatened (T). A taxon likely to become endangered within the foreseeable future throughout all or a significant part of its

range in Kentucky.

Special Concern (S). A taxon that should be monitored because (1) it exists in a limited geographic area in Kentucky, (2) it may become threatened or endangered due to modification or destruction of habitat, (3) certain characteristics or requirements make it especially vulnerable to specific pressures, (4) experienced researchers have identified other factors that may jeopardize it, or (5) it is thought to be rare or declining in Kentucky but insufficient information exists for assignment to the threatened or endangered status categories.

Historical (H). A taxon that has not been reliably reported in Kentucky since 1980 but is not considered extinct or extirpated—see

next designation.

Extinct/Extirpated. A taxon for which habitat loss has been pervasive and/or concerted efforts by knowledgeable biologists to collect or observe specimens within appropriate habitat have failed.

Federal statuses (NMFS 1999; USFWS 1999, 2000) are defined below. Non-breeding birds with a federal status occurring as migrants or visitors in Kentucky (e.g., Charadrius melodus, Mycteria americana) are not included on the list.

Endangered (E). "... any species ... in danger of extinction throughout all or a significant portion of its range ..." (USFWS

Threatened (T). "... any species ... likely to become an endangered species within the foreseeable future throughout all or a significant portion of its range" (USFWS

Proposed Endangered (PE). A taxon proposed

for listing as endangered.

Candidate (C). Taxa for which the USFWS has ". . . sufficient information on biological vulnerability and threats to support proposals to list them as endangered or threatened" (USFWS 1999).

#### DISCUSSION

The list of rare biota includes a lichen and 389 plant and 282 animal taxa considered rare

in Kentucky (Tables 1, 2). Based on generally accepted estimates of the number of native taxa in Kentucky and excluding extinct/extirpated members of each group, the following approximate percent of the groups indicated can be considered Endangered, Threatened, of Special Concern, or Historical: vascular plants—16.5%, gastropods—9.7%, freshwater mussels—42.9%, fishes—26.6%, amphibians and reptiles—28.4%, breeding birds—30.6%, and mammals—21.5%. Although KSNPC continues to refine and expand this list to include new groups, the list does not adequately treat or include several groups of organisms found in Kentucky. The fungi, liverworts, insects, amphipods, isopods, and other groups are important elements of our natural heritage but are poorly known in Kentucky. Researchers are encouraged to continue to gather and publish information about these groups to assist in the evaluation and inclusion of rare taxa on future lists.

Four plants and 42 animals are presumed extinct or extirpated from Kentucky (Tables 2, 3). Most extinct or extirpated animals are freshwater mussels or fishes that have experienced range-wide declines caused by habitat destruction, stream modification, and pollution (Richter et al. 1997). Extirpation and extinction are difficult to prove definitively, so biologists should continue to seek these plants

and animals during field activities. We invite recommendations from knowl-

edgeable individuals regarding native taxa they believe deserve a status change or should be added to or deleted from the list. Each recommendation should include the scientific name of the organism, its habitat requirements, collection information (i.e., localities, number of specimens, dates, disposition of specimens), historical and present distribution, whether the taxon has been specifically sought during field work, threats to its survival, and recommended status. Recommendations should be forwarded to the Director, KSNPC, who will pass the information on to appropriate staff members for timely review and response.

KSNPC intends to publish updated lists in the *Journal* every 4 years. The present lists will be updated annually by submitting a note to the *Journal* listing status and name changes. Interested persons can contact KSNPC for the

Table 1. Endangered, threatened, special concern, and historical biota of Kentucky, 2000.

	Status			Status	
	KSNPC	US		KSNPC	US
Lichens			Ageratina luciae-brauniae	S	
Phaeophyscia leana	E	_	Lucy Braun's white snakeroot	_	
Bottomland lichen			Agrimonia gryposepala	T	_
Plants			Tall hairy groovebur	Т	
Mosses			Amianthium muscitoxicum Fly-poison	1	
Abietinella abietina	Т	_	Amsonia tabernaemontana var.	Т	
Wire fern moss	1		gattingeri		
Anomodon rugelii	T		Eastern blue-star		
A moss			Anemone canadensis	Н	_
Brachythecium populeum	$\mathbf{E}$	_	Canada anemone		
Matted feather moss			Angelica triquinata	$\mathbf{E}$	
Bryum cyclophyllum	$\mathbf{E}$	_	Filmy angelica		
Å moss			Apios priceana	$\mathbf{E}$	T
Bryum miniatum	$\mathbf{E}$	_	Price's potato-bean		
Å moss			Arabis hirsuta var. adpressipilis	$\mathbf{E}$	_
Cirriphyllum piliferum	T	_	Hairy rock-cress		
A moss			Arabis missouriensis	E	_
Dicranodontium asperulum	$\mathbf{E}$	_	Missouri rock-cress		
A moss			Arabis perstellata	T	E
Entodon brevisetus	$\mathbf{E}$	_	Braun's rock-cress		
A moss			Aristida ramosissima	H	_
Herzogiella turfacea	$\mathbf{E}$		Branched three-awn grass		
A moss			Armoracia lacustris	T	
Neckera pennata	T	_	Lake cress		
A moss			Aster acuminatus	T	_
Oncophorus raui	E	_	Whorled aster		
A moss			Aster concolor	T	_
Orthotrichum diaphanum	E	_	Eastern silvery aster	_	
A moss	_		Aster drummondii var. texanus	T	_
Polytrichum pallidisetum	T		Texas aster		
A haircap moss			Aster hemisphericus	E	_
Polytrichum piliferum	E		Tennessee aster		
A haircap moss	_		Aster pilosus var. priceae	T	_
Polytrichum strictum	$\mathbf{E}$	_	White heath aster	_	
A haircap moss	_		Aster pratensis	S	
Sphagnum quinquefarium	E	_	Barrens silky aster		
A peatmoss			Aster radula	$\mathbf{E}$	_
Tortula norvegica	E	_	Low rough aster	TT.	
A tortula			Aster saxicastellii	T	
Vascular Plants			Rockcastle aster	C	
Acer spicatum	E	_	Aureolaria patula	S	
Mountain maple			Spreading false foxglove	C	
Aconitum uncinatum	T		Baptisia australis var. minor	S	_
Blue monkshood	-		Blue wild indigo	c	
Adiantum capillus-veneris	Т	_	Baptisia bracteata var. leucophaea	S	
Southern maidenhair fern	_		Cream wild indigo	Т	
Adlumia fungosa	E		Baptisia tinctoria	1	
Climbing fumitory			Yellow wild indigo	Т	
Aesculus pavia	T	_	Bartonia virginica	1	_
Red buckeye			Yellow screwstem Berberis canadensis	E	
Agalinis auriculata	E	_	American barberry	12	
Earleaf False Foxglove			Berchemia scandens	Т	
Agalinis obtusifolia	$\mathbf{E}$	_	Supplejack		
Ten-lobe false foxglove			Botrychium matricariifolium	E	_
Agalinis skinneriana	E		Matricary grapefern	L	
Pale false foxglove			Botrychium oneidense	E	_
Agastache scrophulariifolia	S	_	Blunt-lobe grapefern	_	
Purple giant hyssop			S-ap		

Table 1. Continued.

	Status			Status	
	KSNPC ·	US		KSNPC	US
Bouteloua curtipendula	S		Carex seorsa	S	_
Side-oats grama			Weak stellate sedge		
Boykinia aconitifolia	T	_	Carex stipata var. maxima	S	
Brook saxifrage			Stalkgrain sedge		
Cabomba caroliniana	T	_	Carex straminea	T	_
Carolina fanwort			Straw sedge		
Calamagrostis canadensis var.	$\mathbf{E}$	_	Carex tetanica	E	_
macouniana			Rigid sedge		
Blue-joint reed grass			Carya aquatica	T	
Calamagrostis porteri ssp. insperata	$\mathbf{E}$		Water hickory		
Reed bent grass			Castanea dentata	E	_
Calamagrostis porteri ssp. porteri	T	_	American chestnut		
Porter's reed grass			Castanea pumila	T	_
Callirhoe alcaeoides	$\mathbf{H}$		Allegheny chinkapin		
Clustered poppy-mallow			Castilleja coccinea	E	_
Calopogon tuberosus	$\mathbf{E}$	_	Scarlet indian paintbrush		
Grass-pink			Ceanothus herbaceus	T	_
Calycanthus floridus var. glaucus	T	_	Prairie redroot		
Sweetshrub			Cheilanthes alabamensis	E	_
Calylophus serrulatus	H		Alabama lip fern		
Yellow evening primrose			Cheilanthes feei	E	_
Carex aestivalis	E	_	Fée's lip fern		
Summer sedge	_		Chelone obliqua var. obliqua	E	
Carex alata	Т	_	Red turtlehead	_	
Broadwing sedge	-		Chelone obliqua var. speciosa	S	_
Carex appalachica	T		Rose turtlehead	J	
Appalachian sedge	•		Chrysogonum virginianum	E	_
Carex atlantica ssp. capillacea	E	_	Greeen-and-gold	L	
Prickly bog sedge	ь		Chrysosplenium americanum	E	
Carex austrocaroliniana	S	•	American golden-saxifrage	L	
Tarheel sedge	J		Cimicifuga rubifolia	Т	
Tarneer seuge Carex buxbaumii	Н		Appalachian bugbane	1	
	11	_		·S	
Brown bog sedge	Н		Circaea alpina	5	
Carex comosa	п		Small enchanter's-nightshade	Т	
Bristly sedge	S		Clematis crispa	1	
Carex crawei	3		Blue jasmine leather-flower	Н	
Crawe's sedge	Т		Coeloglossum viride var. virescens	п	
Carex crebriflora	1 ,		Long-bract green orchis	E	
Coastal Plain sedge	т		Collinsonia verticillata	Е .	
Carex decomposita	T	_	Whorled horse-balm	E	
Epiphytic sedge	Т		Comptonia peregrina	£	
Carex gigantea	1		Sweet-fern	177	,
Large sedge	**		Conradina verticillata	E	
Carex hystericina	Н		Cumberland rosemary	177	
Porcupine sedge	TC		Convallaria montana	E	_
Carex joorii	Ě	_	American lily-of-the-valley		
Cypress-swamp sedge			Corallorhiza maculata	E	
Carex juniperorum	E	_	Spotted coralroot	0	
Cedar sedge	-		Coreopsis pubescens	S	-
Carex lanuginosa	$\mathbf{E}$	_	Star tickseed		
Woolly sedge	-		Corydalis sempervirens	S	_
Carex leptonervia	E	_	Pale corydalis		
Finely-nerved sedge	-		Cymophyllus fraserianus	E	-
Carex reniformis	E	_	Fraser's sedge		
Reniform sedge	_		Cyperus plukenetii	H	-
Carex roanensis	E		Plukenet's cyperus		
Roan sedge			Cypripedium candidum	E	-
Carex rugosperma	T	_	Small white lady's-slipper		
Umbel-like sedge			Cypripedium kentuckiense	S	_
			Kentucky lady's slipper		

Table 1. Continued.

	Status	TIC		Status	
	KSNPC	US		KSNPC	US
Cypripedium parviflorum	T	_	Gentiana flavida	$\mathbf{E}$	_
Small yellow lady's-slipper	**		Yellow gentian	-	
Cypripedium reginae	Н		Gentiana puberulenta	E	-
Showy lady's-slipper	6		Prairie gentian		
Dalea purpurea	S	_	Glandularia canadensis	T	_
Purple prairie-clover	_		Rose verbena		
Delphinium carolinianum	T		Gleditsia aquatica	S	_
Carolina larkspur			Water locust		
Deschampsia cespitosa ssp. glauca	$\mathbf{E}$		Glyceria acutiflora	T	_
Tufted hair grass			Sharp-scaled manna grass		
Deschampsia flexuosa	T		Gnaphalium helleri var. micradenium	H	_
Crinkled hair grass			Small rabbit-tobacco		
Dichanthelium boreale	S	_	Gratiola pilosa	T	_
Northern witch grass			Shaggy hedge-hyssop		
Didiplis diandra	S		Gratiola viscidula	S	
Water-purslane			Short's hedge-hyssop		
Disporum maculatum	S	_	Gymnopogon ambiguus	S	_
Nodding mandarin			Bearded skeleton grass		
Dodecatheon frenchii	S	_	Gymnopogon brevifolius	E	_
French's shooting-star	5		Shortleaf skeleton grass	_	
Draba cuneifolia	E	_	Halesia tetraptera	Т	_
Wedge-leaf whitlow-grass	L		Common silverbell		
Drosera brevifolia	E		Hedeoma hispidum	T	
Dwarf sundew	E		Rough pennyroyal	1	
	Н		Helianthemum bicknellii	T	
Drosera intermedia	п	_		1	
Spoon-leaved sundew	C		Plains frostweed	E	
Dryopteris carthusiana	S		Helianthemum canadense	E	_
Spinulose wood fern	**		Canada frostweed		
Dryopteris ludoviciana	Н	_	Helianthus eggertii	T	7
Southern shield wood fern	_		Eggert's sunflower	~	
Echinodorus berteroi	T	_	Helianthus silphioides	E	_
Burhead			Silphium sunflower	_	
Echinodorus parvulus	$\mathbf{E}$		Heracleum lanatum	E	_
Dwarf burhead			Cow-parsnip		
Eleocharis olivacea ,	S	—	Heteranthera dubia	S	
Olivaceous sedge			Grassleaf mud-plantain		
Elodea nuttallii	T		Heteranthera limosa	S	_
Waterweed			Blue mud-plantain		
Elymus svensonii	S		Heterotheca subaxillaris var. latifolia	T	_
Svenson's wild rye			Broad-leaf golden-aster		
Eriophorum virginicum	$\mathbf{E}$	_	Hexastylis contracta	E	_
Tawny cotton-grass			Southern heartleaf		
Eryngium integrifolium	E	_	Hexastylis heterophylla	S	_
Blue-flower coyote-thistle			Variable-leaved heartleaf		
Erythronium rostratum	S		Hieracium longipilum	T	_
Golden-star			Hairy hawkweed		
Eupatorium maculatum	Н		Houstonia serpyllifolia	E	_
Spotted joe-pye-weed			Michaux's bluets	_	
Eupatorium semiserratum	E		Hydrocotyle americana	E	_
Small-flowered thoroughwort	L		American water-pennywort	_	
Eupatorium steelei	E		Hydrolea ovata	E	_
	Ľ		Ovate fiddleleaf	L	
Steele's joe-pye-weed	T		Hydrolea uniflora	S	
Euphorbia mercurialina	T			J	
Mercury spurge	т		One-flower fiddleleaf	c	
Fimbristylis puberula	T	_	Hydrophyllum virginianum	S	-
Hairy fimbristylis	T.		Virginia waterleaf	**	
a omenta en a la casa train a	T		Hypericum adpressum	H	_
Upland privet	_		Creeping St. John's-wort		
Forestiera ligustrina Upland privet Gentiana decora Showy gentian	S	_	Creeping St. John's-wort  Hypericum crux-andreae  St. Peter's-wort	T	_

Table 1. Continued.

	Status			Statu		
	KSNPC	US		KSNPC	US	
Hypericum nudiflorum Pretty St. John's-wort	Н	_	Lobelia nuttallii Nuttall's lobelia	T	_	
Hypericum pseudomaculatum	H	_	Lonicera dioica var. orientalis Wild honeysuckle	E	_	
Large spotted St. John's-wort	E		Lonicera reticulata	E	_	
Copper iris soetes butleri	E	_	Grape honeysuckle Ludwigia hirtella	E	_	
Butler's quillwort soetes melanopoda	E	_	Hairy ludwigia Lycopodiella appressa	E	_	
Blackfoot quillwort	S		Southern bog club-moss Lycopodiella inundata	E		
uglans cinerea White walnut		_	Northern bog club-moss			
uncus articulatus Jointed rush	S		Lycopodium clavatum Running-pine	E	_	
uncus elliottii Bog rush	Н	_	Lysimachia fraseri Fraser's loosestrife	E	_	
uncus filipendulus Long-styled rush	T	_	Lysimachia radicans Trailing loosestrife	Н	_	
uniperus communis var. depressa	T	_	Lysimachia terrestris	E	_	
Ground juniper Koeleria macrantha June grass	E	_	Swamp-candles  Maianthemum canadense  Wild lily-of-the-valley	T	_	
Krigia occidentalis Western dwarf dandelion	E	_	Maianthemum stellatum Starry false solomon-seal	E	_	
Lathyrus palustris	T	_	Malus angustifolia	S	-	
Vetchling peavine Lathyrus venosus	S		Southern crabapple Malvastrum hispidum Hispid false mallow	T	-	
Smooth veiny peavine eavenworthia exigua var. laciniata	T	<del></del>	Marsĥallia grandiflora	E	_	
Glade cress eavenworthia torulosa	T	_	Barbara's-buttons Matelea carolinensis	E	_	
Necklace glade cress eiophyllum buxifolium	Н	_	Carolina anglepod Melampyrum lineare var. latifolium	T	_	
Sand-myrtle espedeza capitata	S	_	American cow-wheat  Melampyrum lineare var. pectinatum	E	-	
Round-head bush-clover espedeza stuevei	S	_	American crow-wheat Melanthera nivea	S	_	
Tall bush-clover esquerella globosa	T '	C	Snow melanthera Melanthium parviflorum	E .	_	
Lesquereux's bladderpod Lesquerella lescurii	S	_	Small-flowered false hellebore Melanthium virginicum	E	_	
Lescur's bladderpod eucothoe recurva	E	_	Virginia bunchflower Melanthium woodii	· T	_	
Fetterbush  iatris cylindracea	Т	_	False hellebore Minuartia cumberlandensis	E	]	
Slender blazingstar Lilium philadelphicum	Ť.		Cumberland sandwort Minuartia glabra	T		
Wood lily	T		Appalachian sandwort  Mirabilis albida	E		
ilium superbum Turk's cap lily			Pale umbrella-wort			
imnobium spongia American frog's-bit	T	_	Monarda punctata Spotted beebalm	Н	-	
<i>iparis loeselii</i> Loesel's twayblade	T	_	Monotropsis odorata Sweet pinesap	T		
Listera australis Southern twayblade	E	_	Muhlenbergia bushii Bush's muhly	E	-	
Listera smallii	T	_	Muhlenbergia cuspidata Plains muhly	T		
Kidney-leaf twayblade Lobelia appendiculata var. gattingeri Gattinger's lobelia	E		Muhlenbergia glabriflora Hair grass	S	_	

Table 1. Continued.

	Status			Status	
	KSNPC	US		KSNPC	US
Myriophyllum heterophyllum Broadleaf water-milfoil	S		Platanthera cristata Yellow-crested orchid	Т	
Myriophyllum pinnatum Cutleaf water-milfoil	Н		Platanthera integrilabia White fringeless orchid	T	С
Najas gracillima	S		Platanthera psycodes	E	
Thread-like naiad Nemophila aphylla	T		Small purple-fringed orchid  Poa saltuensis	E	_
Small-flower baby-blue-eyes Nestronia umbellula	$\mathbf{E}$		Drooping blue grass  Podostemum ceratophyllum	S	_
Conjurer's-nut Oenothera linifolia	E	_	Threadfoot Pogonia ophioglossoides	E	_
Thread-leaf sundrops Oenothera oakesiana	Н		Rose pogonia Polygala cruciata	E	_
Evening primrose Oenothera perennis	E	_	Cross-leaf milkwort  Polygala nuttallii	Н	_
Small sundrops Oenothera triloba	T		Nuttall's milkwort Polygala paucifolia	E	
Stemless evening-primrose		_	Gaywings		
Oldenlandia uniflora Clustered bluets	E	_	Polygala polygama Racemed milkwort	T _	
Onosmodium molle ssp. hispidissimum	E	_	Polymnia laevigata Tennessee leafcup	E	
Hairy false gromwell Onosmodium molle ssp. molle	E	_	Pontederia cordata  Pickerel-weed	T	_
Soft false gromwell Onosmodium molle ssp. occidentale	E	_	Potamogeton illinoensis Illinois pondweed	S	_
Western false gromwell  Orobanche ludoviciana	Н		Potamogeton pulcher	T	_
Louisiana broomrape			Spotted pondweed Prenanthes alba	E	_
Orontium aquaticum Goldenclub	T	_	White rattlesnake-root Prenanthes aspera	E	
Oxalis priceae Price's yellow wood sorrel	Н	_	Rough rattlesnake-root  Prenanthes barbata	E	_
Parnassia asarifolia Kidney-leaf grass-of-parnassus	E	_	Barbed rattlesnake-root  Prenanthes crepidinea	Т	_
Parnassia grandifolia	E	_	Nodding rattlesnake-root  Psoralidium tenuiflorum	E	
Largeleaf grass-of-parnassus Paronychia argyrocoma	E	_	Few-flowered scurf-pea		
Silvering Paspalum boscianum	S		Ptilimnium capillaceum Mock bishop's-weed	T	_
Bull paspalum Paxistima canbyi	T	_	Ptilimnium costatum  Eastern mock bishop's-weed	S	_
Canby's mountain-lover Pedicularis lanceolata	Н	_	Ptilimnium nuttallii Nuttall's mock bishop's-weed	E	
Swamp lousewort Perideridia americana	Т		Pycnanthemum albescens White-leaved mountain-mint	E	_
Eastern eulophus Phacelia ranunculacea	S		Pycnanthemum muticum	T	_
Blue scorpion-weed			Blunt mountain-mint Pyrola americana	Н	_
Philadelphus inodorus  Mock orange	T	_	American wintergreeen Ranunculus ambigens	S	_
Philadelphus pubescens Hoary mock orange	E	_	Water-plantain spearwort Rhododendron canescens	E	_
Phlox bifida ssp. bifida Cleft phlox	T	_	Hoary azalea Rhynchosia tomentosa	E	_
Phlox bifida ssp. stellaria Starry cleft phlox	T		Hairy snout-bean Rhynchospora globularis	S	
Plantago cordata Heartleaf plantain	Н	_	Globe beaked-rush Rhynchospora macrostachya	E	_
meatical piantalli			Tall beaked-rush	L	

Table 1. Continued.

	Status			Status	
	KSNPC	US		KSNPC	US
Rubus canadensis	E	_	Silene regia	E	_
Smooth blackberry			Royal catchfly		
Rubus whartoniae	T		Silphium laciniatum var. laciniatum	E	
Wharton's dewberry			Compassplant		
Rudbeckia subtomentosa	$\mathbf{E}$		Silphium laciniatum var. robinsonii	T	
Sweet coneflower			Compassplant		
Sabatia campanulata	E	_	Silphium pinnatifidum	S	_
Slender marsh-pink			Tansy rosinweed		
Sagittaria graminea	T		Silphium wasiotense	S	_
Grass-leaf arrowhead			Appalachian rosinweed		
Sagittaria platyphylla	T		Solidago albopilosa	T	T
Delta arrowhead			White-haired goldenrod		
Sagittaria rigida	$\mathbf{E}$		Solidago buckleyi	S	_
Sessile-fruit arrowhead			Buckley's goldenrod		
Salix amygdaloides	H	_	Solidago curtisii	T	_
Peachleaf willow			Curtis' goldenrod		
Salix discolor	H		Solidago gracillima	S	_
Pussy willow			Southern bog goldenrod		
Salvia urticifolia	E		Solidago puberula	S	_
Nettle-leaf sage			Downy goldenrod		
Sambucus racemosa ssp. pubens	$\mathbf{E}$	_	Solidago roanensis	T	
Red elderberry			Roan Mountain goldenrod		
Sanguisorba canadensis	E	_	Solidago shortii	$\mathbf{E}$	E
Canada burnet			Short's goldenrod		
Saxifraga michauxii	T	_	Solidago simplex ssp. randii	S	_
Michaux's saxifrage			Rand's goldenrod		
Saxifraga micranthidifolia	E	_	Solidago squarrosa	H	_
Lettuce-leaf saxifrage			Squarrose goldenrod		
Saxifraga pensylvanica	H	_	Sparganium eurycarpum	E	_
Swamp saxifrage			Large bur-reed		
ichisandra glabra	$\mathbf{E}$	_	Sphenopholis pensylvanica	S	_
Bay starvine			Swamp wedgescale		
Schizachne purpurascens	T	_	Spiraea alba var. alba	E	_
Purple-oat			Narrow-leaved meadowsweet		
Schwalbea americana	H	$\mathbf{E}$	Spiraea virginiana	T	T
American chaffseed			Virginia spiraea		
Scirpus expansus	E	_	Spiranthes lucida	T	_
Woodland beak-rush			Shining ladies'-tresses		
Scirpus fluviatilis	E	_	Spiranthes magnicamporum	T	_
River bulrush			Great Plains ladies'-tresses		
Scirpus hallii	E	_	Spiranthes ochroleuca	S	
Hall's bulrush			Yellow nodding ladies'-tresses		
Scirpus heterochaetus	E	_	Spiranthes odorata	·E	_
Slender bulrush			Sweetscent ladies'-tresses		
cirpus microcarpus	E	_	Sporobolus clandestinus	T	
Small-fruit bulrush	*		Rough dropseed		
Scirpus verecundus	$\mathbf{E}$ .		Sporobolus heterolepis	$\mathbf{E}$	
Bashful bulrush			Northern dropseed		
Scleria ciliata var. ciliata	E	_	Stachys eplingii	$\mathbf{E}$	_
Fringed nut-rush			Epling's hedge-nettle		
Scutellaria arguta	T	_	Stellaria fontinalis	T	_
Hairy skullcap			Water stitchwort		
Scutellaria saxatilis	T	_	Stellaria longifolia	S	_
Rock skullcap			Longleaf stitchwort		
Sedum telephioides	T		Streptopus roseus var. perspectus	E	
Allegheny stonecrop			Rosy twistedstalk		
Sida hermaphrodita	S	_	Symphoricarpos albus	E	-
Virginia-mallow			Snowberry		
Silene ovata	T	_	Talinum calcaricum	E	_
Ovate catchfly			Limestone fameflower		

Table 1. Continued.

	Statu			Status	
	KSNPC	US		KSNPC	
alinum teretifolium	T	_	Vitis labrusca	S	
Roundleaf fameflower			Northern fox grape		
axus canadensis	T	_	Vitis rupestris	T	
Canadian yew			Sand grape		
ephrosia spicata	$\mathbf{E}$		Woodsia appalachiana	$\mathbf{E}$	
Spiked hoary-pea			Mountain woodsia		
haspium pinnatifidum	T		Xerophyllum asphodeloides	Н	
Cutleaf meadow-parsnip			Eastern turkeybeard		
hermopsis mollis	$\mathbf{E}$	_	Xyris difformis	E	
Soft-haired thermopsis			Carolina yellow-eye-grass		
huja occidentalis	T		Zizania palustris var. interior	Н	
Northern white-cedar			Indian wild rice		
orreyochloa pallida	$\mathbf{E}$	_	Zizaniopsis miliacea	T	
Pale manna grass			Southern wild rice		
oxicodendron vernix	· E	_	Animals		
Poison sumac			Gastropods		
ragia urticifolia	$\mathbf{E}$	_	-	т	
Nettle-leaf noseburn			Anguispira rugoderma	T	
repocarpus aethusae	T	_	Pine Mountain tigersnail	C	
Trepocarpus			Antroselatus spiralis	S	
richostema setaceum	$\mathbf{E}$	-	Shaggy cavesnail	C	
Narrow-leaved bluecurls			Appalachina chilhoweensis	S	
rientalis borealis	$\mathbf{E}$	_	Queen crater	C	
Northern starflower			Fumonelix wetherbyi	S	
rifolium reflexum	$\mathbf{E}$	_	Clifty covert	C	
Buffalo clover			Glyphyalinia raderi	S	
rifolium stoloniferum	T	$\mathbf{E}$	Maryland glyph	nr.	
Running buffalo clover			Glyphyalinia rhoadsi	T	
rillium nivale	${f E}$		Sculpted glyph	œ	
Snow trillium			Helicodiscus notius specus	T	
rillium pusillum var. ozarkanum	$\mathbf{E}$	_	A snail	0	
Ozark least trillium			Helicodiscus punctatellus	S	
rillium pusillum var. pusillum	$\mathbf{E}$	_	Punctate coil	0	
Least trillium			Leptoxis praerosa	S	
rillium undulatum	T		Onyx rocksnail		
Painted trillium			Lithasia armigera	S	
riplasis purpurea	Η .	_	Armored rocksnail	0	
Purple sand grass			Lithasia geniculata	S	
lmus serotina	S	_	Ornate rocksnail	0	
September elm			Lithasia salebrosa	S	
Itricularia macrorhiza	$\mathbf{E}$	_	Muddy rocksnail	0	
Greater bladderwort			Lithasia verrucosa	S	
'allisņeria americana	S		Varicose rocksnail	T	
Eel-grass			Mesomphix rugeli	T	
Ternonia noveboracensis	S	_	Wrinkled button	m	
New York ironweed			Neohelix dentifera	T	
Teronica americana	Н	_	Big-tooth whitelip	C	
American speedwell			Patera panselenus	S	
iburnum molle	T	— .	Virginia bladetooth		
Missouri arrow-wood			Pilsbryna sp.	E	
iburnum nudum	E	_	A snail (undescribed)	C	
Possum haw viburnum			Pleurocera alveare	S	
iburnum rafinesquianum var.	T	_	Rugged hornsnail	C	
rafinesquianum			Pleurocera curta	S	
Downy arrowwood			Shortspire hornsnail	ran	
iola septemloba var. egglestonii	S	_	Rabdotus dealbatus	T	
			Whitewashed rabdotus		
Eggleston's violet			$Rhodacme\ elatior$	S	

Table 1. Continued.

	Status			Statu		
	KSNPC	US		KSNPC	US	
Vertigo bollesiana Delicate vertigo	E		Potamilus purpuratus Bleufer	E	-	
Vertigo clappi	E	_	Ptychobranchus subtentum	E	С	
Cupped vertigo Vitrinizonites latissimus	Т	_	Fluted kidneyshell Quadrula cylindrica cylindrica	Т	_	
Glassy grapeskin Webbhelix multilineata	T		Rabbitsfoot Simpsonaias ambigua	T	_	
Striped whitelip Freshwater Mussels			Salamander mussel <i>Toxolasma lividus</i>	E	_	
	E	E	Purple lilliput			
Alasmidonta atropurpurea Cumberland elktoe		Ľ	Toxolasma texasiensis Texas lilliput	E		
Alasmidonta marginata Elktoe	T	_	<i>Villosa fabalis</i> Rayed bean	E	_	
Anodontoides denigratus Cumberland papershell	E	_	Villosa lienosa Little spectaclecase	S	_	
Cumberlandia monodonta Spectaclecase	E	_	Villosa ortmanni	T	_	
Cyprogenia stegaria Fanshell	E	E	Kentucky creekshell Villosa trabalis	E	E	
Epioblasma brevidens	E	E	Cumberland bean Villosa vanuxemensis	T	_	
Cumberlandian combshell Epioblasma capsaeformis	E	E	Mountain creekshell Crustaceans			
Oyster mussel Epioblasma obliquata obliquata	E	E	Barbicambarus cornutus	S		
Catspaw Epioblasma torulosa rangiana	E	E	Bottlebrush crayfish Bryocamptus morrisoni elegans	T	_	
Northern riffleshell Epioblasma triquetra	S	_	A copepod  Caecidotea barri	E	_	
Snuffbox Fusconaia subrotunda subrotunda	S	<u>·</u>	Clifton cave isopod Cambarellus puer	E	_	
Longsolid Lampsilis abrupta	E	E	A dwarf crayfish Cambarellus shufeldtii	S	_	
Pink mucket Lampsilis ovata	E	_	Cajun dwarf crayfish Cambarus parvoculus	E	_	
Pocketbook  Lasmigona compressa	E	_	A crayfish Cambarus veteranus	S	_	
Creek heelsplitter Lasmigona subviridis	Ε.	_	A crayfish Gammarus bousfieldi	E	_	
Green floater Lexingtonia dolabelloides	н	С	Bousfield's amphipod  Macrobrachium ohione	- E		
Slabside pearlymussel			Ohio shrimp			
Obovaria retusa Ring pink	E	Е	Orconectes australis A crayfish	, T	_	
Pegias fabula Littlewing pearlymussel	E	E	Orconectes bisectus Crittenden crayfish	T	_	
Plethobasus cooperianus Orangefoot pimpleback	Ė.	E	Orconectes burri A crayfish	T	-	
Plethobasus cyphyus Sheepnose	S		Orconectes inermis A crayfish	S	_	
Pleurobema clava Clubshell	E	E	Orconectes jeffersoni Louisville crayfish	E	_	
Pleurobema oviforme	E	_	Orconectes lancifer	E	_	
Tennessee clubshell Pleurobema plenum	E	E	A crayfish Orconectes palmeri	E	_	
Rough pigtoe Pleurobema rubrum	E	_	A crayfish Orconectes pellucidus	S		
Pyramid pigtoe  Potamilus capax  Fat pocketbook	E	E	A crayfish  Palaemonias ganteri  Mammoth Cave shrimp	E	E	

Table 1. Continued.

	Status	TIC		Status	
	KSNPC	US		KSNPC	US
Procambarus viaeviridis A crayfish	Т		Pseudanophthalmus horni abditus Concealed cave beetle	T	_
Stygobromus vitreus An amphipod	S		Pseudanophthalmus horni caecus Clifton Cave beetle	T	_
Insects			Pseudanophthalmus horni horni Garman's cave beetle	S	_
Calephelis mutica Swamp metalmark	S	_	Pseudanophthalmus hypolithos	T	
Callophrys irus Frosted elfin	S	_	Ashcamp cave beetle Pseudanophthalmus inexpectatus	T	
Celithemis verna Double-ringed pennant	S		Surprising cave beetle Pseudanophthalmus parvus	T	_
Cheumatopsyche helma	Н	_	Tatum Cave beetle Pseudanophthalmus pholeter	E	_
Helma's net-spinning caddisfly Dryobius sexnotatus	Т	_	Greater Adams Cave beetle Pseudanophthalmus pubescens	T	
Sixbanded longhorn beetle Ephemerella inconstans	Н	_	intrepidus A cave beetle		
An ephemerellid mayfly  Erora laeta	S	_	Pseudanophthalmus puteanus Old Well Cave beetle	T	
Early hairstreak  Euphyes dukesi	S	_	Pseudanophthalmus rogersae Rogers' cave beetle	T	
Duke's skipper Litobrancha recurvata	S	_	Pseudanophthalmus scholasticus	T	_
A burrowing mayfly  Lordithon niger	Н		Scholarly cave beetle Pseudanophthalmus simulans	T	_
Black lordithon rove beetle			Cub Run Cave beetle Pseudanophthalmus tenebrosus	T	_
Lytrosis permagnaria A geometrid moth	E	_	Stevens Creek Cave beetle Pseudanophthalmus troglodytes	T	
Manophylax butleri A limnephilid caddisfly	S	_	Louisville cave beetle Pyrgus wyandot	T	_
Nicrophorus americanus  American burying beetle	Н	E	Appalachian grizzled skipper Raptoheptagenia cruentata	Н	
Ophiogomphus aspersus Brook snaketail	Н	_	A heptageniid mayfly	S	
Ophiogomphus howei Pygmy snaketail	S	_	Satyrium favonius ontario Northern hairstreak		
Papaipema eryngii	<b>E</b> .		<i>Speyeria idalia</i> Regal fritillary	Н	_
Rattlesnake-master borer moth Phyciodes batesii	Н		Stenonema bednariki A heptageniid mayfly	S	-
Tawny crescent Polygonia faunus	Н	_	Stylurus notatus Elusive clubtail	Н	
Green comma Polygonia progne	Н	_	Traverella lewisi A leptophlebiid mayfly	H	_
Gray comma			Fishes		
Pseudanophthalmus audax Bold cave beetle	Т	_	Acipenser fulvescens	E	_
Pseudanophthalmus calcareus Limestone cave beetle	T	_	Lake sturgeon Alosa alabamae	E	С
Pseudanophthalmus catoryctos Lesser Adams cave beetle	E	_	Alabama shad <i>Amblyopsis spelaea</i>	S	_
Pseudanophthalmus conditus Hidden cave beetle	T	_	Northern cavefish  Ammocrypta clara	E	
Pseudanophthalmus desertus major	Т	_	Western sand darter	E	
Beaver cave beetle Pseudanophthalmus exoticus	Н	_	Atractosteus spatula Alligator gar		-
Exotic cave beetle Pseudanophthalmus frigidus	Т	_	Cyprinella camura Bluntface shiner	E	
Icebox cave beetle  Pseudanophthalmus globiceps  Round-headed cave beetle	Т		Cyprinella venusta Blacktail shiner	S	_

Table 1. Continued.

	Status			Statu	
	KSNPC	US		KSNPC	US
Erimystax insignis Blotched chub	E	_	Macrhybopsis gelida Sturgeon chub	Н	С
Erimyzon sucetta	T	_	Macrhybopsis meeki Sicklefin chub	H	С
Lake chubsucker  Esox niger	S	_	Menidia beryllina	T	_
Chain pickerel Etheostoma chienense	E	E	Inland silverside  Moxostoma poecilurum	E	
Relict darter Etheostoma cinereum	S	_	Blacktail redhorse Nocomis biguttatus	S	
Ashy darter Etheostoma fusiforme	E	_	Hornyhead chub Notropis albizonatus	E	E
Swamp darter Etheostoma lynceum	E	_	Palezone shiner Notropis hudsonius	S	_
Brighteye darter Etheostoma maculatum	Т	_	Spottail shiner Notropis maculatus	Т	_
Spotted darter	E		Taillight shiner	E	
Etheostoma microlepidum Smallscale darter		_	Notropis sp. Sawfin shiner (undescribed)		
Etheostoma nigrum susanae Johnny darter	E	. C	Noturus exilis Slender madtom	E	
Etheostoma parvipinne Goldstripe darter	E	-	Noturus hildebrandi Least madtom	E	_
Etheostoma percnurum Duskytail darter	E	E	Noturus phaeus Brown madtom	E	_
Etheostoma proeliare	T	_	Noturus stigmosus	S	
Cypress darter Etheostoma pyrrhogaster	$\mathbf{E}$	_	Northern madtom Percina macrocephala	Т	_
Firebelly darter Etheostoma swaini	E	_	Longhead darter Percina squamata	E	_
Gulf darter Etheostoma tecumsehi	Т		Olive darter Percopsis omiscomaycus	S	_
Shawnee darter Fundulus chrysotus	E	_	Trout-perch Phenacobius uranops	S	_
Golden topminnow Fundulus dispar	E	_	Stargazing minnow Phoxinus cumberlandensis	T	Т
Northern starhead topminnow  Hybognathus hayi	E		Blackside dace Platygobio gracilis	S	
Cypress minnow			Flathead chub		
Hybognathus placitus Plains minnow	S	_	Rhinichthys cataractae Longnose dace	E	
Hybopsis amnis Pallid shiner	Н	_	Scaphirhynchus albus Pallid sturgeon	E	E
Ichthyomyzon castaneus Chestnut lamprey	S	_	Thoburnia atripinnis Blackfin sucker	S	_
Ichthyomyzon fossor Northern brook lamprey	Т	_	Typhlichthys subterraneus Southern cavefish	S	
Ichthyomyzon gagei Southern brook lamprey	$\mathbf{H}_{\uparrow}$ .	_	Umbra limi Central mudminnow	T	
Ichthyomyzon greeleyi Mountain brook lamprey	T	_	Amphibians		
Ictiobus niger	S	Name and Address of the Owner, where the Owner, which is the Owner, where the Owner, which is the Owner, where the Owner, which is the Owner	Amphiuma tridactylum Three-toed amphiuma	E	_
Black buffalo  Lampetra appendix	T		Cryptobranchus alleganiensis alleganiensis	S	_
American brook lamprey Lepomis marginatus	E	_	Eastern hellbender Eurycea guttolineata	Т	
Dollar sunfish Lepomis miniatus	T		Three-lined salamander	T	
Redspotted sunfish Lota lota	S		<i>Hyla avivoca</i> Bird-voiced treefrog	1	
Burbot					

Table 1. Continued.

	Statu			Statu	
	KSNPC	US		KSNPC	
Hyla cinerea	S	_	Ammodramus henslowii	S	-
Green treefrog			Henslow's sparrow		
Hyla gratiosa	S	_	Anas clypeata	$\mathbf{E}$	
Barking treefrog			Northern shoveler		
Hyla versicolor	S	_	Anas discors	T	
Gray treefrog			Blue-winged teal		
Plethodon cinereus	S	_	Ardea alba	$\mathbf{E}$	
Redback salamander			Great egret		
Plethodon wehrlei	$\mathbf{E}$	_	Ardea herodias	S	
Wehrle's salamander			Great blue heron		
Rana areolata circulosa	S	-	Asio flammeus	$\mathbf{E}$	
Northern crawfish frog			Short-eared owl		
Rana pipiens	S		Asio otus	$\mathbf{E}$	
Northern leopard frog			Long-eared owl		
Reptiles			Bartramia longicauda	Н	
-			Upland sandpiper		
Apalone mutica mutica	S	_	Botaurus lentiginosus	H	
Midland smooth softshell			American bittern		
Chrysemys picta dorsalis	T		Bubulcus ibis	S	
Southern painted turtle	_		Cattle egret		
Clonophis kirtlandii	T	_	Certhia americana	$\mathbf{E}$	
Kirtland's snake			Brown creeper		
Elaphe guttata guttata	S	_	Chondestes grammacus	T	
Corn snake			Lark sparrow .		
Eumeces anthracinus anthracinus	T	_	Circus cyaneus	T	
Northern coal skink			Northern harrier		
Eumeces anthracinus pluvialis	$\mathbf{E}$		Cistothorus platensis	S	
Southern coal skink			Sedge wren	_	
Eumeces inexpectatus	S		· Corvus corax	E	
Southeastern five-lined skink			Common raven	L	
Farancia abacura reinwardtii	S		Corvus ossifragus	S	
Western mud snake			Fish crow	5	
Lampropeltis triangulum elapsoides	S	_	Dendroica fusca	Т	
Scarlet kingsnake			Blackburnian warbler	1	
Macroclemys temminckii	T	_	Dolichonyx oryzivorus	S	
Alligator snapping turtle				3	
Nerodia cyclopion	E		Bobolink	E	
Mississippi green water snake			Egretta caerulea Little blue heron	£	
Nerodia erythrogaster neglecta	S	_		E	
Copperbelly water snake	_		Empidonax minimus	£	
Nerodia fasciata confluens	E	_	Least flycatcher	E	
Broad-banded water snake			Falco peregrinus	£	
Ophisaurus attenuatus longicaudus	T	_	Peregrine falcon	TT	
Eastern slender glass lizard	_		Fulica americana	Н	
Pituophis melanoleucus melanoleucus	T	_	American coot	ar.	
Northern pine snake	•		Gallinula chloropus	T	
Sistrurus miliarius streckeri	T		Common moorhen		
Western pigmy rattlesnake			Haliaeetus leucocephalus	E	
Thamnophis proximus proximus	T		Bald eagle	0	
Western ribbon snake	•		Ictinia mississippiensis	S	
Thamnophis sauritus sauritus	S		Mississippi kite		
Eastern ribbon snake	3		Ixobrychus exilis	T	
			Least bittern	_	
Breeding Birds			Junco hyemalis	S	
Accipiter striatus	S	_	Dark-eyed junco		
Sharp-shinned hawk	•		Lophodytes cucullatus	T	
Actitis macularia	E		Hooded merganser		
Spotted sandpiper			Nyctanassa violacea	T	
Aimophila aestivalis	E	_	Yellow-crowned night-heron		
Bachman's sparrow	-		Nycticorax nycticorax	T	
			Black-crowned night-heron		

Table 1. Continued.

	Statu	S		Statu	S
	KSNPC	US		KSNPC	US
Pandion haliaetus	T	_	Mammals		
Osprey			Clethrionomys gapperi maurus	S	_
Passerculus sandwichensis	S	_	Kentucky red-backed vole		
Savannah sparrow			Corynorhinus rafinesquii	S	_
Phalacrocorax auritus	H	_	Rafinesque's big-eared bat		
Double-crested cormorant			Corynorhinus townsendii virginianus	E	· E
Pheucticus ludovicianus	S	_	Virginia big-eared bat		
Rose-breasted grosbeak			Mustela nivalis	S	_
Picoides borealis	E	E	Least weasel		
Red-cockaded woodpecker			Myotis austroriparius	E	_
Podilymbus podiceps	E	_	Southeastern myotis		
Pied-billed grebe			Myotis grisescens	E	E
Pooecetes gramineus	E	_	Gray myotis		
Vesper sparrow			Myotis leibii	T	_
Rallus elegans	E	_	Eastern small-footed myotis		
King rail			Myotis sodalis	E	E
Riparia riparia	S	_	Indiana myotis		
Bank swallow			Nycticeius humeralis	T	_
Sitta canadensis	E	_	Evening bat		
Red-breasted nuthatch			Peromyscus gossypinus	T	
Sterna antillarum	$\mathbf{E}$	E	Cotton mouse		
Least tern			Sorex cinereus	S	
Thryomanes bewickii	S	_	Masked shrew		
Bewick's wren			Sorex dispar blitchi	E	
Tyto alba	S	_	Long-tailed shrew		
Barn owl			Spilogale putorius	S	
Vermivora chrysoptera	T	_	Eastern spotted skunk		
Golden-winged warbler			Ursus americanus	S	
Vireo bellii	S	_	Black bear		
Bell's vireo					
Wilsonia canadensis	S	_			
Canada warbler					

current status of Kentucky's rare organisms. Information about delisted and other taxa are maintained in manual files for use in the event that changes in distribution or status occur.

Each edition of these lists (Branson et al. 1981; Warren et al. 1986; KSNPC 1996, 1997, 1999) has been refined and enhanced with status changes and the addition of new taxonomic groups. As previously noted, these lists are important conservation tools used by KSNPC to focus protection efforts. We hope this information is used by planners and decision makers to conserve Kentucky's unique natural heritage through research, protection, and avoidance.

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Table 2. Diversity and conservation status of the major groups of organisms in Kentucky, 2000.

Number of Kentucky species or taxa <sup>1</sup>	Lichens	Mosses	Vascular plants	Gastropods	Freshwater mussels	Crustaceans	Insects	Fishes	Amphibians	Reptiles	Breeding birds	Mammals
Notive	ımk	326	2262	~ 259	103	unk.	unk.	237	51	52	168	70
vauve Smotio	hunk		745	?	0	unk.	unk.	23	0	_	4	ಬ
NOUC	1	7	370		36	06	48	61	11	18	49	14
KSINFC Montored as nare Vendo Endongoned		15	2.5	g ec	26	i တ	4	28	61	က	18	N
Noint Communication		i rc	116		າດ	ນ	20	13	c1	°C	11	S
KSINI C. Illicateficu VSNPC Special Concern	o C	0	29		4	9	11	16	<u>~</u>	7	16	9
KSNPC Historical	0	0	38	0		0	13	4	0	0	4	0
Presumed Extinct or Extirpated	0	0	4	0	19	0	Τ	œ	0	-	œ	$\mathcal{D}$
Extant/Extirpated												
Threatened	0/0	0/0	10/0	0/0	14/8	1/0	1/0	2/0	0/0	0/0	3/2	3/3
Extant/Extirpated Federally Proposed Endangered												
or Threatened	0/0	0/0	0/0	0/0	0/1	0/0	0/0	0/0	0/0	0/0	0/0	0/0
Fytant Federal Candidate		0	67	0	63	0	0	4	0	0	0	0

<sup>1</sup> Totals include some distinctive subspecies and varieties for gastropods, freshwater mussels, and fishes but only species for lichens, mosses, vascular plants, amphibians, reptiles, breeding birds, and mammals (Mselby 1993); gastropods, freshwater mussels, fishes, amphibians, reptiles, breeding birds, and mammals (KSNPC 2000).

Table 3. Plants and animals presumed extinct or extirpated from Kentucky.

	US Status		US Status
Plants		Fishes	
Caltha palustris var. palustris		Ammocrypta vivax	
Marsh marigold	_	Scalv sand darter	_
Orbexilum stipulatum		Crystallaria asprella	
Stipuled scurf-pea	_	Crystal darter	_
Physostegia intermedia		_ *	
Slender dragon-head		Erimystax x-punctatus	
Polytaenia nuttallii		Gravel chub	_
Prairie parsley		Etheostoma microperca	
Animals		Least darter	_
Freshwater Mussels		Hemitremia flammea	
		Flame chub	_
Dromus dromas	E	Moxostoma lacerum	
Dromedary pearlymussel	£	Harelip sucker	_
Epioblasma arcaeformis		Moxostoma valenciennesi	
Sugarspoon	_	Greater redhorse	
Epioblasma biemarginata Angled riffleshell		Percina burtoni	
Epioblasma flexuosa	_	Blotchside logperch	_
Leafshell		Diotenside logperen	_
Epioblasma florentina florentina	-	Reptiles	
Yellow blossom	E	Masticophis flagellum flagellum	
Epioblasma florentina walkeri	L	Eastern coachwhip	_
Tan riffleshell	E	I	
Epioblasma haysiana	L	Breeding Birds	
Acornshell		Anhinga anhinga	
Epioblasma lewisii		Anhinga	
Forkshell		Campephilus principalis	
Epioblasma obliquata perobliqua		Ivory-billed woodpecker	E
White catspaw	Ε.	Chlidonias niger	-
Epioblasma personata		Black tern	
Round combshell	_		_
Epioblasma propinqua		Conuropsis carolinensis	
Tennessee riffleshell	_	Carolina parakeet	
Epioblasma sampsonii		Ectopistes migratorius	
Wabash riffleshell	_	Passenger pigeon	_
Epioblasma stewardsonii		Elanoides forficatus forficatus	
Cumberland leafshell	· —	Swallow-tailed kite	_
Epioblasma torulosa torulosa		Tympanuchus cupido	
Tubercled blossom	E	Greater prairie-chicken	_
Hemistena lata		Vermivora bachmanii	
Cracking pearlymussel	E	Bachman's warbler	E
Leptodea leptodon			23
Scaleshell	PE	Mammals	
Plethobasus cicatricosus		Bos bison	
White wartyback	. E	American bison	_
Quadrula fragosa		Canis lupus	
Winged mapleleaf	E	Grav wolf	E
Quadrula tuberosa		Canis rufus	3
Rough rockshell	_	9	E
nsects		Red wolf	E
Pentagenia robusta		Cercus elaphus	
Robust pentagenian burrowing mavfly		Elk	_
roogst begrageman outtownik majni,	_	Puma concolor couguar	
		Eastern puma	Ε .

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# First Observations with the Morehead Radio Telescope, Morehead State University, Morehead, Kentucky

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#### ABSTRACT

Herein we report initial astronomical observations made with the Morehead Radio Telescope (MRT). The first radio signals from space were observed with the Morehead Radio Telescope in 1997. The MRT has to date observed a variety of cosmic objects, including galactic sources such as supernova remnants, emission nebula, planetary nebula, extended HI emission from the Milky Way, and the sun, and extragalactic sources such as quasars and radio galaxies. Observations of galactic sources herein reported include Taurus A, Cygnus X, and the Rosette Nebula. Additionally, we report observations of extragalactic phenomena, including Cygnus A, 3C 147, and 3C 146. These initial observations serve as a performance and capability test-bed of the MRT. In addition to the astronomical results of these experiments, tests of the positional accuracy, system sensitivity, and receiver response are inherent in this series of experiments. This paper provides a brief overview of the MRT, including upgrades of major systems, performance characteristics, and a brief discussion of these initial observations.

#### INTRODUCTION

The Morehead Radio Telescope (MRT), Morehead State University, is an instrument designed by faculty and students of Morehead State University and industrial partners to provide a research instrument for undergraduate astronomy and physics students. The MRT also serves as an active laboratory for physics, engineering, and computer science undergraduates and faculty. The telescope operates in the radio regime at a central frequency of 1420 MHz, which corresponds to the hyperfine transition of atomic hydrogen (HI). The HI spatial distribution and flux density associated with cosmic phenomena can be observed with the instrument. The dynamics and kinematics of objects in space can be investigated by observing over a range of frequencies. The sensitivity and versatility of the telescope design facilitate investigation of a wide variety of astrophysically interesting phenomena. The MRT design provides an instrument capable of supporting scientific research in observational astrophysics at radio frequencies. First light was achieved in October of 1997 with routine observations beginning in January 1998. A brief overview of the current MRT instrumentation, description of major subsystems (antenna, alt-azimuth drive and control systems, receiver systems, and controlling computer and interface), is provided that focuses on instrumentation upgrades at the MRT, followed by a discussion of the first observations and results. A more detailed technical overview of the instrument has been previously published (Malphrus et al. 1998). A brief discussion of the instrumentation is included here, as several major systems have evolved beyond the previously described systems.

# The 21 cm Atomic Hydrogen Line

The MRT is designed to operate over a frequency band centered at the 21 cm (1420 MHz) spectral line of atomic hydrogen. Van de Hulst first suggested that the 21 cm line might be detected in the interstellar medium (ISM) in 1945 (Van de Hulst 1945). Unfortunately, Van de Hulst was working in occupied Holland at the time. Searches ensued after

WWII in both the Netherlands and the U.S. The first unambiguous detection of the 21 cm line in the ISM occurred in the U.S. in 1951 and is credited to Ewen and Purcell (Ewen and Purcell 1951). Their experiments were performed with a horn antenna and a sensitive superheterodyne receiver. Subsequently, the HI distribution associated with an ever-increasing number of galactic and extragalactic phenomena have been extensively explored.

The 21 cm emission line arises from a spinparity reversal of hydrogen in the atomic state that corresponds to a transition between the F = 0 and F = 1 hyperfine structures of the electron ground state 12S1/2. A transition can occur between the two hyperfine states because they differ slightly in energy, owing to the interaction between the electron spin and the nuclear spin. When the spin parities align, a higher energy state is achieved at F = 1. The electron spin parity reversal to the lower hyperfine state occurs naturally on the order of once every 10 million years. Atomic collisions in the radiant medium can greatly accelerate this process to the point of producing a continuous emission. Although the emission frequency can be calculated in terms of wellunderstood fundamental constants, a correction for the electron spin g factor must be included. Realization that this anomalous factor must be accounted for resulted when comparisons of the 21 cm line first made in 1947 did not agree with the theoretical value. The discrepancy between the theory and empirical values led to the development of quantum electrodynamics (QED). The currently accepted standard frequency of the 21 cm spectral line is 1420.40575186(30) MHz (Storey et al. 1994). The MRT operates a receiver with a 6 MHz bandwidth centered on this frequen-

The science of cm-wave astronomy has provided significant insight into the structure and evolution of cosmic phenomena and afforded a new perspective of the universe. This perspective is considered significant as roughly three-quarters of the material in the universe exists in the form of hydrogen. The MRT scientific goals are predicated upon low resolution, high sensitivity views of the distribution of HI associated with cosmic phenomena.

## MRT INSTRUMENTATION

The basic design of the MRT includes a wire-mesh parabolic reflecting antenna, alt-azimuth tracking positioner, control and drive systems, receiver and signal processing system, a controlling computer with an interface device, and supporting electronics and hardware. The system is designed around a total power receiver that converts radiation from space concentrated by the antenna system to an electrical signal, which is amplified, modified and interpreted. The basic measurement that the telescope is capable of is an induced antenna temperature, which is translated into an output voltage at the post-detection stage of the back-end receiver. This voltage corresponds to the total integrated flux density intrinsic to the object over the observed frequency band. A detailed technical overview of the instrument has been previously published (Malphrus et al. 1998). A brief discussion of the instrumentation is included here, as several major systems have evolved beyond the previously described systems.

The MRT system is positioned by a nowsecond generation system incorporating a controlling computer, optical isolation system, and robotic drive and control systems developed by MSU faculty and students. The current controlling computer is a 450 MHz Pentium II processor with 128 MB of RAM and 9 GB of hard disk space. A multifunction analog, digital, and timing Input/Output (I/O) data acquisition board is installed in the computer. It contains a 12-bit successive approximation A/ D converter with 16 analog inputs, two 12-bit D/A converters with voltage outputs, 8 lines of transistor-transistor logic compatible I/O, and two counter/timer channels for timing I/O. The board has a 500kS/s single channel sampling rate. The controlling computer positions the telescope, instructs it in robotic tracking of cosmic sources, and controls data collection and storage. The data from a particular experiment are then transferred via ftp to a Sun Sparcstation or high-end PC for imaging and analysis.

# Parabolic Reflecting Antenna

The MRT employs a high-gain  $40 \times 11$  foot antenna designed for L-Band operation. A

surplused Army NIKE-Hercules ANS-17 Radar antenna was obtained and modified for radio astronomy applications. The antenna was selected because of its relatively large aperture, excellent aperture efficiency (afforded by its innovative offset feed design), and low cost. It includes a parabolic reflector, feed horn and waveguide assembly, and azimuth and elevation positioning system. The positioning system provides azimuth coverage of 360° and elevation coverage of now greater than 0-90°. Improvements beyond the previously described system (Malphrus et al. 1998) include the inclusion of a waveguide dehydrator and second-generation digital electronics that drive the positioning system.

# MRT Receiver System

The MRT receiver system design and fabrication program was a joint effort between MSU faculty and Kruth-Microwave Electronics Company of Ellicott City, Maryland. The system design is comprised of single receiver with integral low-noise amplifier directly coupled to the waveguide terminus of the MRT antenna system. The overall receiver system design utilizes a low noise, sensitive, stable receiver to convert the 1420 MHz Hydrogen line frequency to a frequency region suitable for processing by standard laboratory equipment. The system is comprised of two major subsystems—the front and back-end receiver systems. The front-end receiver is coupled to the waveguide terminus, which is mounted on the focal feed support of the MRT superstructure. The MRT front-end receiver system incorporates a GaAs FET low-noise amplifier (LNA) design. The back-end IF receiver consists of a processor with four output signals is housed in the Astrophysics Laboratory Control Room. These output singals consist of 3 RF output ports and one DC detector output. The RF output ports provide accessible signals at 160 MHz (6 MHz bandwidth), 21.4 MHz (6 MHz bandwidth), and 21.4 MHz (2 MHz bandwidth). This strategy permits flexibility in signal processing as evolving experimental needs require. The back-end processor also utilizes a frequency synthesizer, associated power supplies, monitor circuitry, and the controlling computer. The DC voltage is derived from an envelope detector and incorporated into the final stage. Upgrades of the receiver system include a second detector scheme—a digital square law detector, from which the final DC voltage is derived and transmitted to the data acquisition board. All initial observations were made with the MRT receiver system operating in total power mode, using the square law detector.

## Performance Characteristics

The MRT primary system performance characteristics have been empirically measured to assist in understanding the results of astronomical observations. The primary system performance characteristics include system temperature, antenna radiation pattern, and antenna gain. The overall system temperature is measured at a respectable 67.3 K (Kruth 1994). The antenna radiation pattern, which is essentially its directivity function on the sky, has been extensively mapped in 2-D and 3-D to determine the main beam and sidelobe structure. These experiments indicated an elliptical beam pattern with a half-power beamwidth (HPBW) of 0.9° and 3.62° for the major and minor axes respectively (Malphrus et al. 1999). This measurement implies good spatial resolution along the horizontal axis of the antenna and less than ideal resolution along the minor axis. The gain of the antenna has been measured at 41dB, which correlates to good sensitivity to objects with relatively low radio frequency flux.

### Observations

The performance characteristics of the telescope have made possible observations of a variety of cosmic phenomena. Observations of Taurus A, Virgo A, Cygnus A, Cygnus X, the Rosette Nebula, and two quasars—3C 196 and 3C 147—are herein reported. Analysis of the data is very encouraging in that the transit profiles and maximum voltage deflections measured for objects are comparable to values obtained from observations of the same objects on different days and from observations made with other instruments. Reproducibility in the transit profiles of these objects is apparent. In addition, data taken on consecutive days reveal a sidereal shift in the peaks expected as the earth progresses through the ecliptic plane. The observation techniques are described below. Samples of these data are presented in the Results section.

Three basic modes of observation of celestial objects available with the MRT were used in these experiments: transit, tracking, and mapping. Observing celestial objects in transit mode simply involves pointing the telescope due south (180° azimuth), to the appropriate altitude and observing the object as the rotation of the earth moves (apparently) the object through the telescope's field of view (meridian transit). Observing celestial objects in tracking mode involves using the positioning system of the telescope to compensate for the earth's rotation. The computer controls the telescope's motion as it tracks an object as the object (apparently) moves across the sky. Mapping the HI distribution of a radio source involves scanning the telescope's beam across the source's position repeatedly in a manner similar to the raster scan process utilized to produce television images. Each observational mode is described in some detail below.

## Transit Observations

After a target object is selected, the position of the object must be determined as well as the time of meridian transit. Some understanding of coordinate systems is essential to these astronomical observations. The sky operates on the Equator Coordinate System. The MRT operates on the Horizon Coordinate System (as the antenna is alt-azimuth mounted). Therefore, an object's celestial position must be converted to local horizon coordinates to be observed. The values, then, that are important for observing in transit mode are the object's altitude (converted from declination) and local sidereal time of meridian transit.

Determination of transit times in L.S.T. For the given date and zone time (Z.T.) the local sidereal time (L.S.T.) can be calculated for the observer's longitude. L.S.T. is the local hour angle (L.H.A.) of the vernal equinox, that is, the right ascension of the observer's meridian. To calculate L.S.T. from E.S.T. is non-trivial. The following is one of numerous algorithms that may be used:

$$\begin{array}{lll} \text{L.S.T.} = & \text{K.} & + & 1.0027379(\text{UT}) & + \\ & & (0.06570982 \times \text{D}) - \text{Longitude/} \\ & & 15^{\circ} \end{array}$$

where:

UT = Universal time in decimal hours(UT + 5 hours = E.S.T.)

D = Number of days since December 31 of the previous year

Longitude = Geographic west longitude (83° 26<sup>m</sup> for Morehead, KY)

K = 17.369382 (1998); 17.385297 (1999); 17.401211 (2000)

The transit time (time at which an object crosses one's local meridian) may be determined from the object's hour angle. The relationship between sidereal time, hour angle, and right ascension of an object is given:

$$H.A. = S.T. - R.A.$$

Transit occurs when the object crosses the local meridian, i.e., when the H.A. = 0; therefore transit occurs when the L.S.T. equals the R.A. of the object. Transit times may be determined by, calculating the E.S.T. of an object when L.S.T. equals the objects R.A. Alternately, an object achieves meridian transit when the L.S.T. is equal to the object's right ascension. Using the Sidereal clock (designed and constructed by students at the Astrophysics Laboratory) to determine transit time simplifies this process.

Determination of transit altitudes. To record the voltage profile of an object at meridian transit, one must know the object's altitude (a) above the local horizon. To find the altitude, one must know the hour angle (H), the declination ( $\delta$ ) of the object, and the geographic latitude of the telescope ( $\varphi$ ) must be known. The hour angle of an object at meridian transit by definition is 0. Transit altitudes were calculated using the following equation:

$$\begin{array}{l} a \, = \, \sin^{-1}((\sin \, \delta)(\sin \, \varphi) \\ + \, (\cos \, \delta)(\cos \, \varphi)(\cos \, H)) \end{array}$$

The values of L.S.T. transit and transit altitude are required to position the telescope to observe an object in transit mode. Values obtained for the celestial objects observed are given in Table 1.

### Instrumental Procedures

After the coordinate conversion was complete, the MRT Operator program was invoked and the telescope was driven to the ap-

Table 1. Coordinates and flux density values for the initial objects observed with the Morehead Radio Telescope, Morehead State University, Morehead, Kentucky by Malphrus et al. from 1997–2000. Right Ascension and Declination represent fixed values while transit altitude was calculated.

Cosmic object	Right ascension	Declination <sup>2</sup>	Transit time (LST)	Transit altitude	RF flux (Jy) <sup>3</sup> (20 cm)
Virgo A (M87)	12h 28m 18s	+12° 40 <sup>m</sup>	12h 28m 18s	64.4°	970
Taurus A	05h 31m 30s	+21° 58 <sup>m</sup>	05h 31m 30s	73.7°	1,420
Rosette Nebula	06 <sup>h</sup> 29 <sup>m</sup> 18 <sup>s</sup>	+04° 57 <sup>m</sup>	06 <sup>h</sup> 29 <sup>m</sup> 18 <sup>s</sup>	56.7°	105
3C 147	05h 38m 43.2s	+49° 49.6 <sup>m</sup>	05h 38m 43.2s	78.4°	58
3C 196	08h 10m 00.1s	+48° 22 <sup>m</sup>	$08^{\rm h}\ 10^{\rm m}\ 00.1^{\rm s}$	79.8°	59
Cygnus A	19h 57m 45s	+40° 36 <sup>m</sup>	$19^{\rm h}\ 57^{\rm m}\ 45^{\rm s}$	87.4°	8,100
Cygnus X	20 <sup>h</sup> 19 <sup>m</sup> 36 <sup>s</sup>	$+40^{\circ}~06^{\circ}$	$20^{\rm h}~19^{\rm m}~36^{\rm s}$	87.4°	410
Cygnus B	20h 48m 12s	+29° 30 <sup>m</sup>	20h 48m 12s	82.3°	252

<sup>1.2</sup> Right Ascension and Declination values are taken from the NASA/IPAC Extragalactic Database (NED). NED is operated by the Jet Propulsion, Laboratory, California Institute of Technology, under contract with the National Aeronautics and Space Administration.

<sup>3</sup> RF fluxes at 20 cm reported are taken from the Third Cambridge Catalog (3C) produced by the Cavendish Laboratory of Cambridge University, UK.

propriate position for each object. Pre- and post detection gains were set (on the back-end receiver IF processor and square law detector, respectively) based upon published values for the flux density at 20 cm for each of the observed sources (Kraus 1986). These values are reported in Table 1. for each astronomical source. Each source was observed for approximately one hour to provide baseline data before and after each meridian transit. The procedures followed for tracking and mapping runs involve the same initial procedures as those for transit observations, but also include additional, more elaborate procedures described below.

# **Tracking Observations**

Tracking observations involve continuously calculating the object's azimuth and elevation as time progresses and driving the telescope to the calculated position. The telescope can also be "manually" driven from the back-end with the drive and control system to maintain "peak on source". A combination of using the computer-generated coordinates and nudging the telescope with the manual drive system has proven most effective thus far. All peak voltages herein reported were measured during tracking observations utilizing a variety of integration times.

# Mapping Observations

Observing with the MRT in mapping mode is the most challenging but rewarding observational procedure. A pre-determined area (usually rectangular) is scanned with the telescope beam as the object is tracked across the sky. The target area is scanned in a manner similar to the raster scan process utilized by monitors and television sets to produce images. A single eleveation is scanned over a predetermined range of azimuths in one direction (i.e., CW). The telescope is then driven to a lower elevation (typically 1°) and the same range of azimuth values is scanned by driving the telescope in the opposite (CCW) direction. This process is repeated until the entire target area is scanned. 2-D (contour) and 3-D (topographic) maps of the HI associated with the astronomical object can be produced with these data. These maps represent the spatial distribution of HI in the target region (integrated over the entire receiver post-detection bandwidth). The data reduction and imaging software utilized in these experiments is the HiQ package developed by National Instruments. Additional software that allows the observer to reduce and edit the data as well as transform the raw data into an appropriate matrix format was developed by MSU students. A more detailed description of the mapping procedure, including the details of data reduction, is described in a later publication.

### RESULTS

To date, observations of a variety of cosmic phenomena have been undertaken. Herein, we describe observations of Virgo A, the Cygnus A Complex, Taurus A, the Rosette Nebula, 3C 196, and 3C 147. Telescope receiver

Cosmic object	Pre-detec- tion gain	Post-detec- tion gain	Integra- tion time (s)	TOS	$\Delta V_{\scriptscriptstyle max}$	$N_{\rm mis}$	S/N
Virgo A (M87)	40%	900x	15	2160s	4.1v	0.3v	13.6/1
Taurus A	40%	900x	10°	864s	4.9v	0.1v	49/1
Rosette Nebula	40%	1000x	15	684s	5.7v	0.05v	114/1
3C 147	40%	1000x	<b>1</b> s	1200s	3.8v	1.5v	2.5/1
3C 196	40%	1000x	<b>1</b> s	1200s	5.8v	0.3v	19.3/1
Cygnus A	50%	900x	1s	2400s	2.5v	0.1v	25/1
Cygnus X	50%	900x	1s	1080s	0.5v	0.1v	5/1
Cygnus B	50%	900x	1s	2400s	0.3v	0.1v	3/1

Table 2. Telescope settings and results obtained for the initial astronomical observations made with the Morehead Radio Telescope, Morehead State University, Morehead, Kentucky by Malphrus et al. from 1997–2000.

settings, integration times, and other observational parameters such as time on source are listed in Table 2. Also listed are induced voltages, and calculated signal-to-noise ratios. Predetection gain, post-detection gain, and integration time are telescope values set by the observer. Time on source (TOS) is determined by the spatial extent of the object when observed in transit mode.  $V_{max}$ ,  $N_{min}$ ,  $\Delta V$ , and S/N represent experimental results. V<sub>max</sub> represents the maximum or peak voltage induced by the cosmic object and is proportional to the flux density of the object as explained below. N<sub>rms</sub> is the baseline width, which also corresponds to the rms noise temperature of the system.  $\Delta V$  is the peak voltage minus the averaged baseline voltage level.

The peak voltage  $\Delta V$ , the basic measurement in total power mode is a function of the induced antenna temperature ( $\Delta T$ ). The antenna and front- and back-end receivers of a radio telescope system together act as a radiometer for measuring the temperature of distant regions of space coupled to the system through the radiation resistance of the antenna. The temperature of the radiation resistance is determined by the temperature of the emitting region seen by the beam of the antenna as defined by its directivity function, i.e., its radiation pattern. The temperature of the antenna radiation resistance is referred to as induced antenna temperature. A mathematical expression describing induced antenna temperature demonstrates that the induced temperature is a function of the cosmic radiator convolved with the antenna's directivity function (Kraus 1986):

$$\Delta V \approx k\Delta T = w$$

$$= \frac{1}{2} Ae \int \int B(\theta, \, \phi) Pn(\theta, \, \phi) \, d\Omega$$

where:  $\Delta V$  = induced post-detection voltage, volts

 $\Delta T$  = induced antenna temperature, K

w = received power per unitbandwidth,  $w \times Hz^{-1}$ 

 $A_e$  = effective aperture (physical aperture × aperture efficiency),  $m^2$ 

 $B(\theta, \phi)$  = source brightness distribution, dimensionless

 $Pn(\theta, \phi)$  = antenna radiation pattern, dimensionless

 $d\Omega = \sin\theta \, d\theta \, d\phi = \text{element of solid angle, rad}^2$ 

 $k = \text{Boltzman's constant } 1.38 \times 10^{-23} \text{ J} \times \text{K}^{-1}$ 

Given that the cosmic object's energy flux density is also a function of the cosmic radiator's brightness convolved with the antenna's directivity function (Kraus 1986)

$$\Delta S = \iint\limits_{Source} B(\theta, \phi) Pn(\theta, \phi) \ d\Omega,$$

where:  $\Delta S = \text{total source flux density, w} \times m^{-2} \times Hz^{-1}$ 

 $B(\theta, \phi) = \text{source brightness distribution, dimensionless}$ 

 $Pn(\theta, \phi)$  = antenna radiation pattern, dimensionless

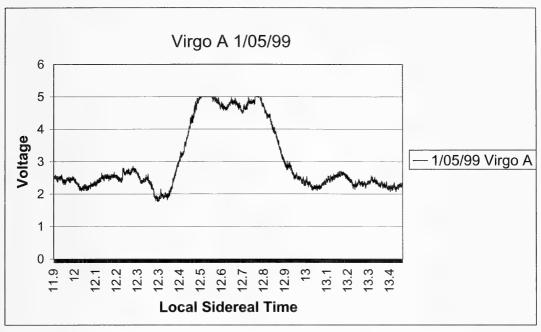


Figure 1. Morehead Radio Telescope, Morehead State University, Morehead, KY. A transit observation of Virgo A taken with the MRT on January 5, 1999.

 $d\Omega = \sin\theta \ d\theta \ d\phi = \text{element of}$ solid angle, rad<sup>2</sup>

It follows that  $\Delta S$ simplifies to

$$\Delta S = \frac{2k\Delta T}{Ae}.$$

A relative value as opposed to an absolute value for  $\Delta T$ , and therefore  $\Delta S$ was obtainable during these experiments.  $\Delta T$  can be calibrated against a standard induced voltage or astronomical flux calibrator in future experiments. The  $S/N_{rms}$ , however, can be measured from the voltage profiles because  $\Delta V_{max}$  corresponds to the signal (S), and  $N_{rms}$ , the rms noise corresponds to the baseline width. The calculated S/N values for each observation are given in Table 2. The S/N is expressed as a ratio. A variety of graphical representations of the data as well as a brief description of the astronomical objects observed are provided below.

# Virgo A

Virgo A is the fifth brightest radio object in the sky, with a flux density of 970 Jy. The object is a radio galaxy with some strange features that make it highly visible in the radio spectrum. Virgo A has an odd jet that extends from the west-side of the nucleus. This jet, apparent at extremely high spatial resolutions, is thought to be associated with material ejected from the nucleus. The system is an extremely powerful emitter of both waves and X-rays. A transit observation of Virgo A taken with the MRT on 5 Jan 1999 is provided in Figure 1.

## Taurus A

Taurus A is a supernova remnant (SNR) in the constellation Taurus; it is also known as the Crab Nebula. Taurus A is located about 6300 light years away and is the fourth brightest radio object in the sky. It has a flux density of 1420 Jy. Successive transit observations of Taurus A were made on 19–20 Aug 1997. These observations are provided in Figure 2. An expected sidereal shift of 4 minutes of time in the transit peaks is observed.

### The Rosette Nebula

The Rosette nebula is a supernova remnant (SNR) within our galaxy that has some inter-

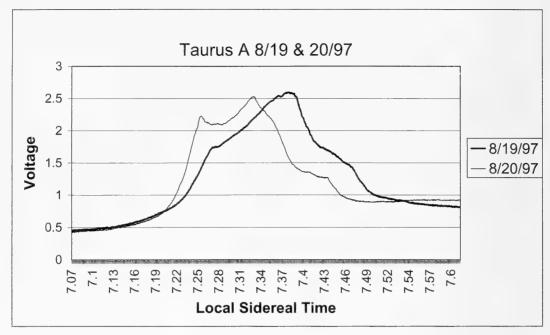


Figure 2. Morehead Radio Telescope, Morehead State University, Morehead, KY. Successive transit observations of Taurus A made on August 19th and 20th, 1997.

esting features. It is also classified as an emission nebula—a cloud of high temperature gas. The atoms in the cloud are energized by ultraviolet light from a nearby star and emit radiation as they fall back into lower energy states; hence we can see the Rosette Nebula in the radio regime. Emission nebulae are usually sites of star formation; the Rosette Nebula does in fact have star formation occurring in its outer regions. A transit observation made of the Rosette Nebula is shown in Figure 3.

## 3C 147

3C 147 (also known as [HB89] 0538  $\pm$  498 and QSO OG  $\pm$ 465) is a distant quasar that has been previously observed by numerous investigators. The quasar is extremely distant and faint in the optical spectrum exhibiting a visual magnitude of 17.8 and a redshift of z = 0.54500 79 (NASA/IPAC Extragalactic Database (NED), 1999). It is optically variable and unresolved by most instruments. The object shows an unusually complex, nonlinear structure that varies with time. Superluminal sep-

aration of two components in the core region has been observed. A jet is embedded in the diffuse emission region. VLA images at 1 GHz indicate a weak component north of the main component opposite the jet with respect to the core. Although the object is spatialy unresolved by the MRT, its luminous output in the radio spectrum is well above the detection limits of the instrument. Figure 4 depicts a transit observation of 3C 147 in which a favorable signal to noise ratio of 2.5:1 was achieved.

## 3C 196

3C 196 (also known as [HB89] 0809+483) is a distant quasar in the constellation Lynx. It is an extremely distant and faint object, exhibiting a redshift of z=0.87100 and an apparent visual magnitude of 17.79 79 (NASA/IPAC Extragalactic Database (NED) 1999). The object has a famous and well-studied absorption system at z=0.43685, which gives rise to a host of metal line species. The 21 cm absorption is especially significant because it occurs in a resolved background source, which allows useful

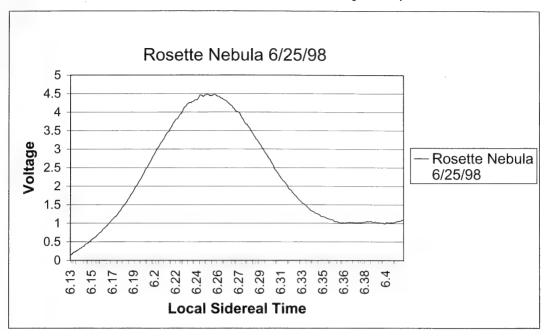


Figure 3. Morehead Radio Telescope, Morehead State University, Morehead, KY. A transit observation of the Rosette Nebula taken with the MRT on June 25, 1998.

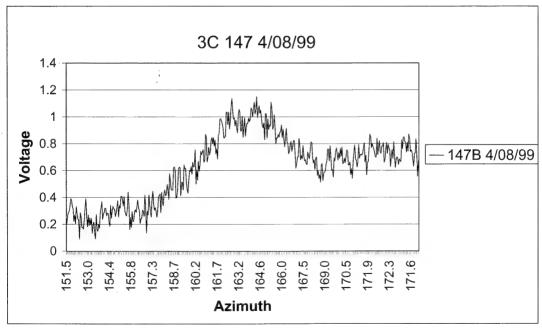


Figure 4. Morehead Radio Telescope, Morehead State University, Morehead, KY. A scan observation made of the quasar 3C 147 on April 8, 1999.

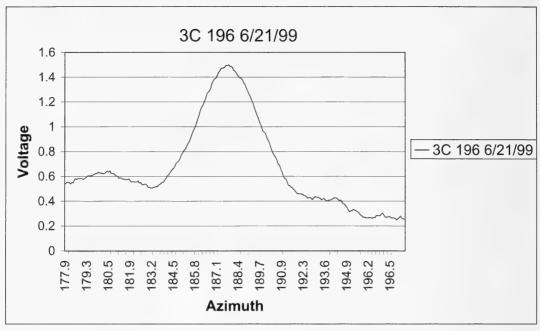


Figure 5. Morehead Radio Telescope, Morehead State University, Morehead, KY. A scan observation made of 3C 196 on June 21, 1999.

limits to be placed on the absorber size. There is also an associated absorber in this object at z=0.8714 with an apparent infall velocity of  $100~\rm kmsec^{-1}$ . A transit observation made with the MRT of 3C 196 is presented in Figure 5.

# The Cygnus A Complex

The Cygnus A Complex, a very active region in the radio sky, contains at least three major components, Cygnus A, Cygnus B, and Cygnus X. Cygnus A is a distant radio galaxy approximately 1 billion light years away. It is one of the best-known radio sources in the sky but it has no bright visible object that corresponds to the radio emission. No visible object was associated with the radiation until 1951 when astronomers at the Palomar Observatory found an object that appeared as a pair of unresolved 18th magnitude objects. Cygnus A is the second most luminous radio object that is observable, with a flux density of 8100 Jy, second only to Cassiopeia A. Cygnus A represents the largest peak in the topographical representation of the data. Cygnus X, the second strongest peak in the data, is associated with

the famous Cygnus X black hole. The radio waves emanate from the accretion disk associated with the black hole. The accretion disk is produced as the black hole's gravity well accretes matter from its companion star, swirling it into a flattened disk and heating the infalling material to 106 K, causing it to radiate in the X-ray and radio regions. Cygnus X is considered among the most convincing candidates for a galactic black hole. Since Cygnus X lies close to the line of sight of Cygnus A, it is almost impossible to observe one object and not the other in a transit observation. Since these objects lie close to each other in line of sight, but not in physical space, their combined observable radiation is called the Cygnus A Complex. We interpret the component labeled Cygnus B in the data to be associated with the Cygnus Loop, an ancient supernova remnant in the galaxy some 770 pc distant from earth. A transit profile of the Cygnus complex as well as three other perspectives of the HI distribution as produced with the MRT is shown in Figure 6. Complex structural de-

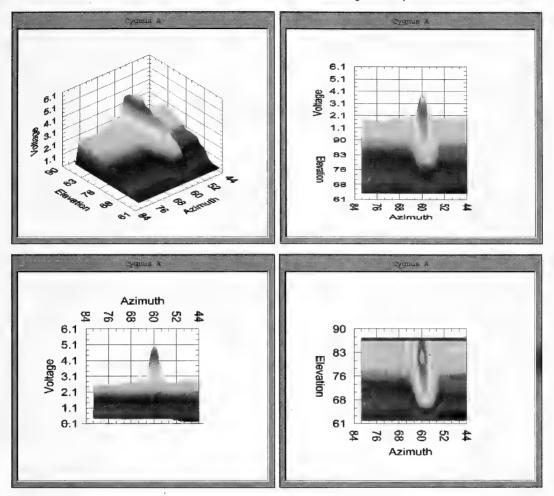


Figure 6. Morehead Radio Telescope, Morehead State University, Morehead, KY. Four perspectives of a map of the spatial distribution of HI associated with the Cygnus A, B, and X radio sources.

tail is evident in the HI associated with these regions.

## DISCUSSION

Results of these initial observations are very promising. The HI profiles and distributions compare favorably to transit profiles and HI distribution maps produced with comparable instruments. Transit profiles of the Cygnus A Complex and Virgo A are extremely similar in structure to ones produced with the NRAO Green Bank 40-foot Radio Telescope and the University of Indianapolis 5-meter antenna. The HI distribution map compares very fa-

vorably to those produced with the NRAO Green Bank 40-ft. Radio Telescope and the Ohio State University "Big Ear." These HI profiles and maps compare favorably to previously produced images despite the MRT's rather elliptical beam.

Results of these initial observations are encouraging in that they attest to the performance characteristics of the MRT. The reproducibility apparent in the data attests to the MRT receiver stability and pointing accuracy. The results of these initial experiments are very exciting in that the capabilities of the instrument to perform more extensive and per-

spicacious experiments is realized. Challenges still exist, however, in the procedural elements of each observing mode. The results of observations in transit mode are haunted by the elliptical, vertically-oriented beam. Circular beams are ideal for these type experiments. Observations made in scanning mode are challenging in that it is difficult to scan the telescope at a constant rate and produce the same number of data points for each scan. Reducing the data and preparing the data matrix for 3-D maps is complicated by this problem. In tracking mode, there are the inherent problems of pointing accuracy. Even still, these early observations are encouraging and indicate that the MRT systems have, even at this formative stage, exceeded expectations.

A limitation of the current data is that the observations are essentially unclaibrated. Future experiments may attempt to "bracket" observations of the astronomical sources with observations of known flux calibrators, a strategy utilized at the National Radio Astronomy Observatory's Very large Array (VLA). Other possibilities include injecting a noise source of a standard voltage directly into the waveguide to coaxial transition via a hardware (calibration) switch. This strategy is employed with the National Radio Astronomy Observatory's 40-foot Radio Telescope. Still another possibility involves generating a test (calibration) signal with an RF generator in the Laboratory Control Room and coupling it to a quasi-directional test antenna. Observations of the astronomical sources could then be bracketed by observations of the calibration signal.

## **Next Experiments**

A next generation of experiments is indicated as a result of analysis of these early data. MRT systems experiments to be performed include determining the electrical focus of the antenna system, determining the mechanical focus of the antenna system, empirically determining the minimum detectable flux density, and mathematically modelling the antenna surface. The future, in terms of the possibilities of astronomical observations, is very exciting. The instrument is capable of observing many of the most exotic and energetic astronomical phenomena in the universe. Astronomical investigations include observations of

a wider variety of cosmic sources (including such galactic objects as black holes, pulsars, supernova remnants, starbirth nebulae, and extragalactic objects such as active galaxies, radio galaxies, and interacting galaxy systems), the production of a map of the HI distribution associated with the Milky Way, an all-sky northern hemisphere map, and SETI projects.

### ACKNOWLEDGMENTS

The Morehead Radio Telescope Project was funded by the National Science Foundation under the Instruments and Laboratory Improvements Program, NASA and by Morehead State University. The MRT receives continued support from Morehead State University's College of Science and Technology, Department of Physical Sciences Department of Industrial Education and Technology and from Kruth-Microwave Electronics of Hanover, Maryland, and Dan Puckett Engineering, Morehead, Kentucky. This research has made use of the NASA/IPAC Extragalactic Database (NED), which is operated by the Jet Propulsion Laboratory, California Institute of Technology, under contract with the National Aeronautics and Space Administration.

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# Agrimonia (Rosaceae) in Kentucky with Notes on the Genus

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### ABSTRACT

The genus Agrimonia (Rosaceae) consists of about 15 species worldwide, 7 in the U.S., and 4 in Kentucky: A. gryposepala, A. parviflora, A. pubescens, and A. rostellata. The reported presence in Kentucky of three additional species—A. eupatoria, A. microcarpa, and A. striata—could not be verified. Included are a key to and descriptions of the Kentucky taxa and notes on biology and uses of Agrimonia.

## INTRODUCTION

The genus Agrimonia (Rosaceae), with about 15 species, occurs in North America, South America, Eurasia, and South Africa. Because of the problems of identification of some species of the genus and the uncertainty concerning which species actually occur in Kentucky, we decided to examine the Kentucky species to determine their taxonomy and distribution. Additionally we began a survey of previous work on Agrimonia, presenting here stray notes—not an exhaustive literature review—on the genus.

## AGRIMONIA IN KENTUCKY

Species of Agrimonia—the agrimonies were early recorded as components of the Kentucky flora. M'Murtrie (1819), in his Florula Louisvillensis, listed three: A. eupatoria (a European taxon, almost certainly a misidentification for one of the native Kentucky species), A. parviflora, and A. sylvatica (the last mentioned name we have as yet been unable to trace). Short, Peter, and Griswold's (1833) catalog of the phaenogamous [sic] plants of Kentucky—which, so far as we are aware, is the earliest attempt to account for the flora of a U.S. state—has two: A. eupatoria and A. parviflora. The following year Short and Peter (1834) changed A. parviflora to A. suaveolens (the latter now regarded as a synonym of the former). According to Medley (1993), Short's specimens labelled A. eupatoria and deposited at PH have been annotated as A. striata, an identification we regard as suspect. Kentucky specimens collected in the 1830s by either Short or Peter and sent to us on loan from PH are A. parviflora, A. pubescens, and A. rostellata; A. striata was not represented. Price (1893) had only A. eupatoria for Warren

County; almost certainly this too is a misidentification. Half a century later McFarland (1942) listed the four species that we recognize as representing the genus in Kentucky; Braun (1943) listed only three, not including A. gryposepala. Browne and Athey (1992) listed seven species for the state: the four we recognize in our paper and three others—A. eupatoria, A. microcarpa, and A. striata—the occurrence of which in Kentucky we have not been able to confirm; the listing of these three is, we believe, based on misidentification. In the most recent summary of the Kentucky flora Medley (1993) accepted for the state A. gryposepala, A. microcarpa, A. parviflora, A. pubescens, and A. rostellata.

## MATERIAL AND METHODS

The data herein are based on 421 herbarium specimens (ca. 12% misidentified) of Kentucky Agrimonia borrowed from 14 herbaria; on the 46 collections of Agrimonia we and colleagues made from 1996 through 1999 in 18 counties of Kentucky (specimens in KNK); and on previously published accounts of the genus (Bicknell 1896, 1901a, 1901b; Bush 1916; Fernald 1950; Gleason and Cronquist 1991; Kline and Sørensen 2000; K.R. Robertson 1974; Robinson 1900; 1901; Rydberg 1913; Skalický 1973; Svenson 1941; Torrey and Grav 1838–1840). From herbarium sheets we recorded the following data: county, location, collector(s), date; leaf length; leaflet count, length, and width; sepal length; hair length; and fruit length and width ("fruit" = fruiting hypanthium with enclosed achene(s) but excluding bristles and sepals).

When collecting specimens of *Agrimonia*, one should be certain to obtain the roots, which are helpful in identification, and, if pos-

sible, mature, fully reflexed fruits that are at least half mature.

We saw no specimens of Agrimonia from 35 of Kentucky's too many (120) counties: Adair, Boyd, Carroll, Christian, Clay, Cumberland, Elliott, Fulton, Graves, Grayson, Hancock, Harrison, Henderson, Hopkins, Johnson, Knott, Knox, Leslie, Livingston, Logan, Martin, Mason, Mercer, Metcalfe, Monroe, Owsley, Perry, Russell, Scott, Shelby, Simpson, Trimble, Union, Washington, and Webster.

## **TAXONOMY**

In North America Agrimonia can easily be distinguished from other, sympatric genera of the Rosaceae by its herbaceous habit, its five yellow petals, its five sepals, and its hypanthium ("fruit") armed with hooked bristles. To identify Agrimonia correctly, one must learn to distinguish the different types of stem vestiture (see Jain and Singh [1973] for a study of the hairs of A. eupatoria): (1) minute gland-tipped hairs, the stalk short, few-celled; (2) long, straight or curved, eglandular hairs 1.0–4.0 mm; and (3) short, often curved, sometimes matted, eglandular hairs < 0.5 mm.

Our species of *Agrimonia* flower from July to August or September. Individuals are rare to frequent, but never, in our experience, common or abundant.

Figures 1 and 2 are of Agrimonia parviflora, a representative member of the genus.

# Agrimonia Linnaeus

Herbs perennial, erect, hemicryptophytic, with crystals of calcium oxalate in parenchymatous tissues (Murata and Umemoto 1983). Roots fibrous, sometimes with fusiform tubers. Stems branched or unbranched. Leaves alternate, stipulate, odd 1-pinnate, of 2 main sizes, major (larger) leaflets 3–19(21), toothed, terminal leaflet often the largest, minor (smaller) leaflets 0-37, sometimes bractletlike, interspersed among major leaflets. Inflorescence racemose, terminal and often axillary, few to many flowered; flowers nearly sessile, pedicels bracteate at base, ascending, stipe spreading or reflexed in fruit. Flowers subopposite to alternate, 4-9 mm in diameter, bibracteolate, perigynous; sepals 5, persistent, forming a beak in fruit; petals 5, yellow (very rarely white); stamens 5-20; pollen 3-colporate; ovaries 2, enclosed by hypanthium, styles exserted. **Fruit** (i.e., fruiting hypanthium plus enclosed achene[s] but excluding bristles and sepals) constricted at the throat, indurate, hemispheric to top-shaped, stipitate, rim bearing erect to reflexed, hooked bristles; achenes 1 or 2. Type species: *Agrimonia eupatoria* L. For illustrations of this species see Phelouzat (1963) and Ross-Craig (1956).

# Key to Kentucky Species of Agrimonia

The l:w data in the key and in each description below refer to the average length:width ratio (plus range of values) of the blade of the largest terminal leaflet on a plant.

- 1. Major leaflets mostly leaves 3–9; l:w < 3.0
  - 2. Rachis of inflorescence copiously pubescent, eglandular or nearly so or the glands concealed by hairs; leaflets downy pubescent beneath; roots with fusiform tubers ...... A. pubescens

- 1. **Agrimonia gryposepala** Wallr. [Greek *grypos*, curved, and New Latin *sepalum*, sepal] Figures 3, 7, 11

**Herb** 3–18 dm, roots without tubers. **Stem:** vestiture of 2 types: (1) minute, gland-tipped hairs; and (2) long, stiff hairs to 3.2 mm. **Leaves:** to 21 cm; major leaflets 3–9, ovate or obovate to elliptic or rhombic, 1– $10.5 \times 0.8$ –5.2 cm, adaxially glabrous or nearly so or with a few scattered long hairs, abaxially with



Figure 1. Agrimonia parviflora, a representative species of the genus,  $\times$  ½. From Zardini 1971, with permission.

gland-tipped hairs and with long hairs 0.5-2 mm; 1:w=2.0 (1.8-2.5); minor leaflets 0-9, to 2.3 cm, 0-3 pairs between major leaflets. **Inflorescense** abundantly glandular and with sparse, long, stiff hairs to 2 mm. **Fruit** (excluding bristles sepals)  $6.8-7.5\times3.5-5.8$  mm, glandular, the hypanthium top-shaped to campanulate, deeply grooved, the ridges, base, and pedicel with a few scattered, stiff, white hairs to 0.5 mm, the grooves glandular, otherwise glabrous; sepals in fruit 2-3 mm; bristles 2.5-3.7(4.0) mm, in 4-5 rows, lowermost row sharply reflexed. 2n=56 (Brittan 1953).

Woodland margins, thickets, clearings, fields, and disturbed sites.

Agrimonia gryposepala is rare and of limited occurrence in Kentucky. Harlan is the only Kentucky county from which we have seen a herbarium specimen to document the occurrence of the species in the state.

Agrimonia gryposepala is considered a threatened species in Kentucky (KSNPC 1996). Kline and Sørensen (1990) discussed lectotypification and synonymy of this species.

2. Agrimonia parviflora Sol. In Ait. [Latin

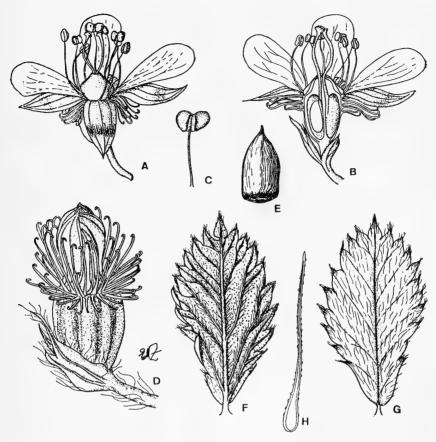


Figure 2. Agrimonia parviflora. Details of flower and leaflet. (A) Flower with 2 forward petals removed,  $\times$  10. (B) Longitudinal section through flower, with bracteoles,  $\times$  10. (C) Stamen,  $\times$  15. (D) "Fruit," with bracteoles,  $\times$  10. (E) Achene,  $\times$  10. (F) Leaflet, adaxial surface,  $\times$  2. (G) Leaflet, abaxial surface,  $\times$  2. (H) Trichome from abaxial surface of leaflet,  $\times$  20. From Zardini 1971, with permission.

parvus, small, and flos, flower, alluding to size of the flowers] Figures 1, 2, 4, 8, 12, 15

**Herb** 5–20 dm; roots without tubers. **Stem:** vestiture of 3 types, each sometimes sparse: (1) minute, gland-tipped hairs; (2) long,  $\pm$  stiff hairs to 3.5(4) mm; and (3) short hairs < 0.5 mm. **Leaves:** to 28 cm; major leaflets (5)9–19(21), lanceolate or oblanceolate to narrowly elliptic (rhombic), 1–8.5  $\times$  0.6–2.5 cm, adaxially minutely pubescent, abaxially with copious gland-tipped hairs and with long hairs 1–3 mm; l:w = 4.0 (3.3–4.8); minor leaflets 15–43, to 2.5 cm, 1–4 pairs between major leaflets. **Inflorescence** minutely glandular, with sparse, short hairs < 0.5 mm and long, stiff, more or less straight or curved hairs to 2 mm. **Fruit** (excluding bristles and sepals) 2.3–

 $4.0(5.0) \times 2.5$ –4.0(4.5) mm, glandular, the hypanthium top-shaped to campanulate, shallowly grooved, the ridges, base, and pedicel often with a few scattered, stiff, white hairs to 0.5 mm, the grooves glandular, otherwise glabrous; sepals in fruit 1.5–1.7 mm; bristles 1.0–3.0 mm, in 3–4 rows, lowermost row spreading to ca. 90° or reflexed. 2n=28 (Hara and Kurosawa 1968).

Moist or wet soil at swamp or stream edges, grassy areas, meadows, thickets, and roadside ditches; sometimes in dry, open places.

Agrimonia parviflora is widely distributed in Kentucky. The documented county-distribution of the species in the state is as follows: Allen, Bath, Bell, Bourbon, Breckinridge, Butler, Calloway, Campbell, Casey, Clark, Edmonson, Fleming, Floyd, Franklin, Garrard,

Greene, Hardin, Harlan, Jackson, Jefferson, Kenton, Laurel, Lee, Lincoln, Lyon, Madison, Magoffin, Marion, McCracken, McCreary, Meade, Menifee, Montgomery, Morgan, Muhlenberg, Pike, Powell, Pulaski, Rockcastle, Rowan, Trigg, Warren, Whitley, and Wolfe.

A study of the reproductive success and breeding system of *A. parviflora* was recently completed by Brann (1998) (see under Biology: Pollination below).

3. **Agrimonia pubescens** Wallroth [Latin *pubis*, downy] Figures 5, 9, 13

**Herb** 3–16 dm; roots with fusiform tubers. **Stem:** vestiture of 2 or 3 types: (1) minute, gland-tipped hairs, these sparse or sometimes lacking; (2) long, stiff hairs to 3 mm, these often sparse; and (3) short hairs < 0.5 mm; types 2 and 3 not always clearly distinguished. **Leaves:** to 24 cm; major leaflets (3)5–9(13), lanceolate or oblanceolate to oblong or elliptic or sometimes obovate,  $1.4-10.7 \times 2-4.9$  cm, adaxially sparsely pubescent, abaxially downypubescent, the hairs 1–2 mm; l:w = 2.2 (1.7– 3.0); minor leaflets 4–11, to 1.7 cm, 1–3 pairs between major leaflets. **Infloresence** minutely glandular, the glands often sparse or even lacking, with short hairs < 0.5 mm and long, stiff hairs to 2 mm, the long hairs sometimes sparse or even lacking. Fruit (excluding bristles and sepals)  $4.0-5.5(6.0) \times 2.3-4.0$  mm, glandular (sometimes sparsely so), the hypanthium top-shaped to campanulate, shallowly to deeply grooved, the ridges, base, and pedicel with a few scattered, stiff, hairs 0.5-0.8 mm, the grooves often with a strip of conspicuous to inconspicuous white, upwardly appressed hairs <0.5 mm (rarely seemingly white farinose); sepals in fruit 1.5–1.7 mm; bristles 1.0– 3.2 mm, in 3–4 rows, lowermost row spreading

Dry to mesic woodlands, woodland edges, disturbed sites.

Agrimonia pubescens is widely distributed in Kentucky. The documented county-distri-

bution of the species in the state is as follows: Anderson, Barren, Boone, Boyle, Bracken, Breathitt, Breckinridge, Bullitt, Caldwell, Campbell, Clark, Clinton, Edmonson, Estill, Fayette, Floyd, Franklin, Gallatin, Green, Greenup, Hardin, Harlan, Hart, Henry, Jackson, Jefferson, Jessamine, LaRue, Laurel, Letcher, Lincoln, Lyon, Madison, Marshall, McLean, McCreary, Meade, Menifee, Muhlenberg, Nelson, Nicholas, Ohio, Oldham, Owen, Pendleton, Pike, Pulaski, Robertson, Todd, Trigg, Warren, Wayne, Wolfe, and Woodford.

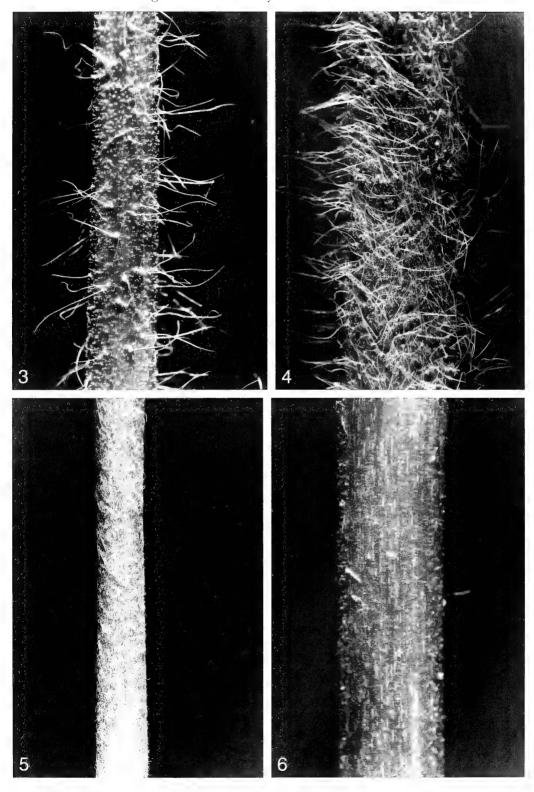
4. **Agrimonia rostellata** Wallroth [Latin *rostellum*, beak, alluding to the connivent sepals on the fruit] Figures 6, 10, 14

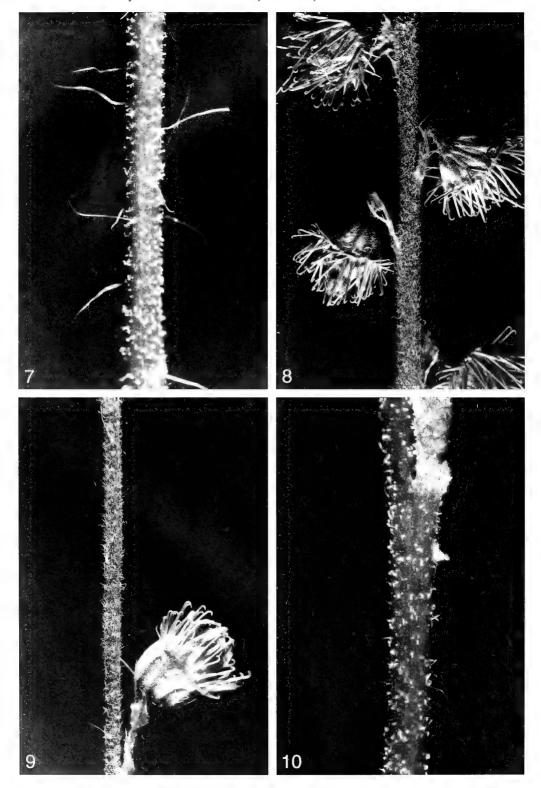
**Herb** 2–10 dm; roots with fusiform tubers. **Stem:** vestitute of 3 types: (1) minute, glandtipped hairs, these sparse to abundant; (2) long, stiff hairs to 2.5(3) mm, these sometimes sparse; and (3) sparse, short hairs < 0.5 mm. Leaves: to 24 cm; major leaflets 3-9(11), obovate to broadly elliptic,  $2.5-10.5 \times 1.5-5.6$ cm, adaxially glabrous or sometimes with a few long hairs, abaxially with copious gland-tipped hairs and with long hairs  $\pm 1$  mm; l:w = 1.9 (1.6-2.5); minor leaflets 0-8, to 2.5 cm, 1(0-2) pair between major leaflets. **Inflorescense** sparingly to copiously glandular, with sparse, short hairs < 0.5 mm and long, stiff hairs to 1 mm, both types sometimes absent. Fruit (excluding bristles and sepals)  $3.2-4.0 \times 2.0-$ 3.0 mm, obscurely to conspicuously glandular, otherwise glabrous or sometimes with a few short hairs <0.2 mm at base or on the pedicel, the hypanthium hemispherical, shallowly grooved; sepals in fruit 1.5-1.8 mm; bristles 1.7-2.0 mm, in 3-4 rows, lowermost row spreading to ca. 90°.

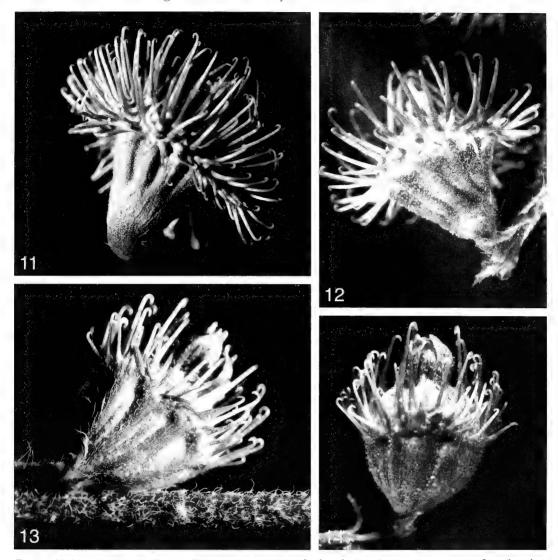
Dry to mesic woodlands, woodland edges, disturbed sites.

Agrimonia rostellata is widely distributed in Kentucky. The documented county-distribution of the species in the state is as follows:

Figures 3–6. Agrimonia. Median portions of stems, showing vestiture. Figure 3, A. gryposepala (stem diam. 2.5 mm). Figure 4, A. parviflora (stem diam. 3.5 mm). Figure 5, A. pubescens (stem diam. 2 mm). Figure 6, A. rostelllata (stem diam. 2 mm).







Figures 11–14. Agrimonia. "Fruits." Figure 11, A. gryposepala (length 7 mm). Figure 12, A. parviflora (length 3 mm). Figure 13, A. pubescens (length 4.5 mm). Figure 14, A. rostelllata (length 3.2 mm).

Allen, Anderson, Ballard, Barren, Bath, Bell, Breathitt, Breckinridge, Bullitt, Caldwell, Calloway, Campbell, Carlisle, Carter, Clark, Clinton, Crittenden, Daviess, Edmonson, Estill, Fayette, Floyd, Garrard, Grant, Greenup, Hardin, Hart, Hickman, Jefferson, Jessamine, Kenton, LaRue, Laurel, Lawrence, Letcher, Lewis, Lyon, Madison, McCreary, McLean, Meade, Menifee, Morgan, Nelson, Nicholas, Oldham, Pike, Powell, Pulaski, Robertson,

Figures 7–10. Agrimonia. Median portions of inflorescence axis, showing vestiture. Figure 7, A. gryposepala (axis diam 1.5 mm). Figure 8, A. parviflora (axis diam. 2 mm). Figure 9, A. pubescens (axis diam. 2 mm). Figure 10, A. rostelllata (axis diam. 1 mm).

Rockcastle, Rowan, Spencer, Taylor, Todd, Trigg, Warren, Wayne, and Woodford.

### EXCLUDED SPECIES

Agrimonia eupatoria L.

Agrimonia eupatoria has been ascribed to Kentucky several times starting with M'Murtrie (1819), but no documenting Kentucky specimens are known to us. The species may not even be naturalized in North America (Kline and Sørensen 1990) in spite of the many reports that it is so. It was, however, recorded in 1924 as a wool-waste plant from Massachusetts (Weatherby 1924), which may explain, in part, Fernald's (1950) attributing it to "waste places and old fields, local, Mass., Wisc. and Minn." (We have seen century-old Minnesota specimens at MU labelled as A. eupatoria; they are A. gryposepala.) Gleason and Cronquist (1991) wrote merely that the species is "occasionally intr. In our range." Kartesz and Meacham (1999) ascribed the species, in eastern North America, to three Canadian provinces and eight U.S. states (but not Kentucky); and, in the west, to one province and two states; most of these reports are based on literature records, not on first-hand study of specimens. But Kline and Sørensen (n.d.), in their account of Agrimonia for Flora of North America, excluded the taxon from the FNA area: "Agrimonia eupatoria, a European species, has been sporadically cultivated in the flora area. We can find no evidence that this introduced species, which rarely escapes cultivation, has become an established element of the flora." Earlier, Bicknell (1896) reached the same conclusion: "... the true Agrimonia Eupatoria is not known at all as an American plant. . . . "

Of A. eupatoria Weatherby (1924) wrote that "it resembles our native A. gryposepala in that the stem and the rachis of the inflorescence are clothed with minute glandular puberulence mixed with long, non-glandular hairs. It is readily recognized, however, by its commonly more compact habit, the lower internodes of the stem tending to be short, thus bringing the leaves close together, by its generally smaller leaflets, and by the characters of the fruiting calyx. The body of the mature hypanthium is rather narrowly top-shaped and measures from the base to the point of insertion of the hooked bristles about 5 mm. In A. gryposepala the corresponding measurement is about 3 mm."

With respect to Kentucky, the species is one that could have been—or is—cultivated here in medicinal plant or other gardens. But we know of no evidence for such cultivation. (Seeds said to be of A. eupatoria are available from several websites.)

In 19th century U.S. floristic literature the accounts of Agrimonia were hopelessly confused as to both nomenclature and taxonomy (Bush 1916), with "A. eupatoria" being often a catch-all name for any U.S. agrimony except the distinct A. parviflora. Clarification of the nomenclature and taxonomy of the U.S. representatives of the genus began in the last years of the 19th century and the early years of the 20th century (Bicknell 1896, 1901a, 1901b; Britton and Brown 1897; Robinson 1900, 1901; Robinson and Fernald 1908; Rydberg 1913). Some authors, though, are still somewhat confused; we hide their identity to protect the guilty.

The 19th century confusion in Agrimonia is best illustrated by the accounts of the genus published in the 1890s in two major floristic works for northeastern U.S. The sixth edition of Gray's Manual (Gray 1890) includes only two species of the genus—A. eupatoria and A. parviflora. The first edition of the Britton and Brown Illustrated flora (Britton and Brown 1897) includes six species, excluding A. eupatoria and commenting that "The European A. Eupatoria L. differs markedly in foliage and fruit from any of our species." Following the lead of the Illustrated flora, the 7th edition of Gray's manual (Robinson and Fernald 1908) included six species but not A. eupatoria. The most recent edition of the Manual (Fernald 1950) includes seven, A. eupatoria having been reinstated—probably in error—as a member of the flora.

Agrimonia microcarpa Wallr.

This species has been ascribed to Kentucky in several works (e.g., Browne and Athey 1991; Fernald 1950; Greenwell 1935; Kearney 1893; Medley 1993; Radford et al. 1968). Even a county of occurrence—Bullitt—has been mentioned. We made an unsuccessful attempt to locate confirming specimens. According to Kline and Sørensen (1990), this

southeastern species ranges north to North Carolina, Tennessee, and Missouri.

Agrimonia microcarpa would key to A. pubescens in the key above. Although recognized as a distinct taxon by various authors, its distinguishing characters seem to us to be rather weak, overlapping in most cases with those of A. pubescens; it has been considered a variety of that species (Ahles 1964; Radford et al. 1968). To separate A. microcarpa from A. pubescens the following features have been among those used: stem hairs 3-4 mm (m) vs.1-3 mm (p); major leaflets 3-7 (m) vs. 3-13(p); major leaflets broadly rounded distally (m)vs. acute to obtuse (p); stipules  $\pm$  falcate to  $\pm$ half round and deeply incised (m) vs.  $\pm$  broadly half ovate, incised but not deeply (p); and mature hypanthium with a distinct rim and as broad as long (m) vs. mature hypanthium with a obsolete rim and usually longer than broad (p).

# Agrimonia striata Michx.

Agrimonia striata is a northern and western species ranging south, in eastern U.S., to Iowa, Illinois, Michigan, Ohio, West Virginia, Pennsylvania, and New Jersey (Kline and Sørensen 1995). Its roots are slender (non-tuberous); it has (3)5–9(11) major leaflets; its abaxial leaflet surfaces are sparingly pubescent, the hairs to 1 mm and confined mostly to veins; its inflorescence is glandular and pubescent, the hairs 1–2 mm; and its hypanthium grooves have a strip of minute, appressed hairs. Voss (1985) pointed out that the stipules of its mid-cauline leaves usually have a prolonged lanceolate terminal tooth or lobe.

### BIOLOGY

Biological data on Agrimonia, at least on our North American species, are relatively scarce. The best sources of such data on the European species known to us are the accounts of Agrimonia in two editions of Hegi (1923, 1995). Only A. eupatoria, long used medicinally in Europe and elsewhere, has been studied with any degree of thoroughness.

### Pollination

Few observations on pollination of agrimonies are available. The species, at least in Europe, bears pollen flowers, which do not produce nectar and are visited by pollen-eating insects. "Flies and bees are attracted to the slender spikes of flowers [of A. eupatoria] by a scent reminiscent of apricots" (RDA 1981).

The pollination mechanism of A. eupatoria was described thus by Knuth (1908):

At the base of the two styles in this species there is a fleshy ring that looks like a nectary, though no secretion has been observed. The 5–7 stamens on the margin of this disk attain the same level as the stigmas, and their anthers dehisce laterally. The anthers incline inwards, and therefore come into contact with the stigmas. The individual flowers bloom for a single day only, and open very early in the morning. The stamens, which are at first divergent, bend inwards in the course of the day, until they touch one another and the stigmas. Comparatively few insects visit the flowers, but these may bring about either cross- or self-pollination. From the above description it is clear that the latter occurs automatically, and it is obviously effective.

Palynological data on Agrimonia (i.e., A. eupatoria) are included in Hegi (1995; light microscope and SEM photos of pollen); Erdtman (1952; description of pollen); and Reitsma (1966; line drawings of pollen).

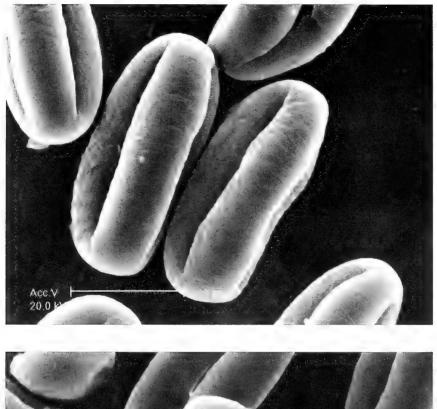
The pollen of A. eupatoria was described thus by Erdtman (1952): "3-colpor(oid)ate (constricticopate), prolate (48  $\times$  31  $\mu$ ). Sexine thicker than nexine, tegillate; endosexine considerably thinner than ectosexine. The latter finely striate."

We include, as Figure 15, a photomicro-

graph of pollen of A. parviflora.

Mueller (1883) recorded as pollinators of European A. eupatoria the following: Diptera–Syrphidae, nine species representing six genera (Ascia, Eristalis, Melanostoma, Melithreptus, Rhyngia, Syritta); Muscidae, one species (Anthomyia); and Hymenoptera–Apidae (a "small" species of Halictus). To these Knuth (1908) added a species of Syrphus (Diptera: Syrphidae) and a species of Apis, one of Bombus, and one of Prosopis (Hymenoptera: Apidae). Clapham et al. (1987) mentioned "Diptera and Hymenoptera."

North American data on pollination of agrimonies are even fewer than those from Europe. Indeed, we have found only two reports: C. Robertson (1928), working near Carlinville, Illinois, recorded a species of *Chloralictus* (Hymenoptera: Halictidae) as a visitor to *A. striata*; and Macroberts and Macroberts (1997) wrote that "captured pollinators [of *A.* 



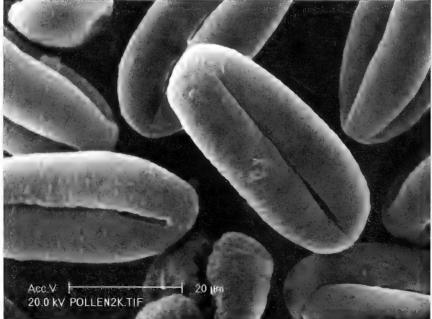


Figure 15. Agrimonia parviflora. Pollen grains,  $\times$  2000. (Voucher: Thieret 52394, Rowan County, Kentucky, 29 Aug 1980 [KNK]). Photomicrograph by Brenda K. Racke.

incisa, a southeastern U.S. species] were small bees of the subfamily Halictinae (Hymenoptera, Apoidea, Halictidae), all probably the same species."

In a study of the breeding system of A. parviflora, Brann (1998) found the species to be self-compatible. Its flowers opened by 0900 and closed by 1400 and were open for only 1 day. Once a flower opened, the availability of its pollen for dispersal and the receptivity of its stigmas were synchronous.

# Dispersal

The fruits of Agrimonia, with their hooked bristles, are dispersed by animals, including humans, to the fur or clothing of which they readily become attached (Coffey 1993; Keville 1991; Macroberts and Macroberts 1997; RDA 1981; Ridley 1930; S.M. Robertson 1973). Dispersal even by birds on rare occasions might be possible as suggested by the following quote from Swink and Wilhelm (1994): "The senior author once rescued a Goldfinch that was hopelessly trapped in the prickly fruits of [A. gryposepala]." Macroberts and Macroberts (1997) suggested that many of the fruits, though seemingly well adapted for longrange dispersal, probably simply drop near the parent stem. Such in-situ dispersal, it seems to us, would result in close groups of plants, but—except in one instance, a group of ca. 20 individuals of A. parviflora—the agrimonies we have seen in Kentucky occur as scattered plants.

Dispersal by water, at least for streamside plants, is presumably possible, the fruits being able to float for about a week, at least in the case of *A. eupatoria* (Ridley 1930).

# **Tuberous Roots**

Half of the North American species of Agrimonia (A. incisa, A. microcarpa, A. pubescens, and A. rostellata) have tuberous roots the function of which is unknown. Macroberts and Macroberts (1997) suggested that "all of these species might occur in fire-dependent or droughty areas where food reserves or the alternatives of clonal reproduction might be important." Perhaps water storage is important, too: Our Kentucky species, except often A. parviflora, grow in dry—sometimes impressively dry—sites.

Cytology

The base chromosome number of Agrimonia is x = 7 (Darlington and Wylie 1955; Hegi 1995; Iwatsubo et al. 1993). The U.S. species with reported counts are A. parviflora, a tetraploid (2n = 28; Hara and Kurosawa 1968), A. gryposepala, an octoploid (2n = 56; Brittan 1953), and A. striata, a tetraploid or octoploid (2n = 28, 56; Hegi 1995). Agrimonia in Japan includes species at the 4×, 6×, and 8× levels. In Eurasia, A. eupatoria is a tetraploid (2n = 28) and A. pilosa is an octoploid (Hegi 1995; Löve and Löve 1961). A hybrid, A. eupatoria × A. procera, is a hexaploid (2n = 42) as is A. nipponica × A. pilosa var. japonica (Iwatsubo et al. 1993).

### **MEDICINAL**

The best-known species of Agrimonia is the Eurasian A. eupatoria. In older herbal literature it was almost panacean, being used in a number of ways for an impressive array of ills from A (asthma) almost to Z (warts) (Hegi 1995). In recent literature (e.g., Bartram 1995; Bown 1995; Duke 1985, 1997; Foster and Duke 2000; Keville 1991; Ody 1993; Schauenberg and Paris 1977; Swanston-Flatt et al. 1990; Wood 1997) it is said to have been used to treat a diversity of ailments including amenorrhea, bed-wetting, conjunctivitis, cystitis, diabetes, diarrhea, eczema, gallstones, gout, hemorrhoids, incontinence, laryngitis, migraines, nosebleed, rheumatism, skin inflammation, sore throats, and ulcers. A somewhat more restrained assessment of the virtues of the species (LRNP 1995) concludes that "it does appear to have justifiable use as a mild antiseptic and topical astringent" but cautions that "internal uses of this herb require further verification." Commission E approves A. eupatoria to treat diarrhea, inflamed mucous membranes of the mouth and throat, and mild, superficial inflammation of the skin (Blumenthal 1998). The astringent properties of A. eupatoria presumably derive from its high tannin content: roots, 25.8%; rhizome, 16.4%; leaves, 16%; and stem, 5.8% (Hegi 1995).

Wood's (1997) 27-page discussion of the medicinal and other virtues of *Agrimonia* is the most detailed and wide-ranging that we have seen.

A tea made from A. eupatoria has been

used to treat dysentery (Anonymous 1856). The plant, in a mixture of powdered frogs and human blood, was once "recommended for all internal hemmorrhages" (Johnson 1862). Hartwell (1982), in a survey of plants used against cancer, included many data from historical literature on *A. eupatoria*.

Langer and Kubelka (1998) described rhizome anatomy of A. eupatoria in comparison to that of Potentilla erecta (Radix Tormentil-

lae), another medicinal plant.

Within the past 2 decades or so, A. pilosa, another Eurasian species, has been increasingly written about, e.g., the effect of a water extract of the plant on tumors (Miyamoto et al. 1987) and acute pulmonary thrombosis (Hsu et al. 1987). In Chinese herbal medicine A. pilosa is used to expel tapeworms (Nigg and Seigler 1992, and "Agrimonia" is one of the herbs that "regulate" the blood (Ehling and Swart 1996).

The mention of *A. eupatoria* in older literature on North American medicinal plants may or may not refer to this European species, which is quite similar to some of our indigenous taxa of the genus. The plant may well have been introduced early to the continent as a medicinal plant (S.M. Robertson 1973), but problems with identification remain. Erichsen-Brown (1989) referred these early records unequivocally to the native *A. striata*. Why she chose this species and not any of the several other indigenous ones is not explained; her selection is another unverifiable datum.

Agrimonia was early noted in reports of the uses of American medicinal plants by Europeans. In the "first systematic publication concerning the American materia medica" (Lloyd in Smith 1812), that of Johann David Schoepf (1787), A. eupatoria is described as having astringent, roborant, prophylactic, diuretic, and vulnerary properties; the name A. eupatoria is probably a misidentification for one of the North American taxa. Cutler (1785), in his work on some of the "vegetable productions" of New England, wrote of Agrimonia (no specific epithet given but presumably a native species) as growing "by fences" and being useful to treat fevers and jaundice. Smith (1812), in his "dispensatory" on Ohio Valley plants, described "agrimoney" (again no species indicated) as "a native of the woods [and thus certainly an indigenous species], but friendly to cultivation." Its virtues included being useful as a tonic and in diabetes, involuntary emission of urine, and dysentery and "other fluxes." Rafinesque (1828) called A. eupatoria a "mild astringent, tonic, and corborant" and noted its use for diarrhea, dysentery, "relaxed bowels," and asthma. He included a color illustration in his account of the species, but this is unidentifiable to species. His description of the range and habitat of the plant— "The Agrimonia Eupatoria is spread from Canada to Missouri and Carolina, and grows in woods, fields, glades and near streams"—is obviously based on an indigenous species. In several Shaker communities (1847-1874) A. eupatoria was "highly recommended in bowel complaints, gravel, asthma, coughs, and gonorrhea." Again, that this "A. Eupatoria" was a native U.S. species is made almost certain by the statement that it was "found by the roadsides and borders of fields. Can. and U.S." (Miller 1976).

Eastern and central North American Native Americans included at least two species of Agrimonia—A. gryposepala and A. parviflora—among the plants they used medicinally (Erichsen-Brown 1989; Moerman 1998). Snakebite, jaundice, diabetes, nosebleed, urinary troubles, nephritis, and diarrhea were among the problems for which the agrimonies were taken; they were also used—conflictingly, it seems to us—as an antidiarrheal medication and as an emetic, though probably not simultaneously.

"Agrimony" (presumably A. eupatoria) is one of the the "Twelve Healers" divinely revealed to the British physician Edward Bach, M.D. (Harrar and O'Donnell 1999). Bach wrote of this herb in his system of healing with flower essences: "An herb . . . for cheerful, peaceful people who loathe discord and whose jovial exterior covers inner torment, sometimes resulting in problems with drugs or alcohol."

Various "Agrmonia" websites list sources of seeds (A. eupatoria, A. pilosa) and medicinal preparations and uses including even a homeopathic remedy from A. eupatoria.

### **MISCELLANEOUS**

Although the species of Agrimonia, as we know them in Kentucky, are anything but "weedy," at least A. gryposepala can, on oc-

casion, be troublesome. Of this species Muenscher (1955) wrote: "badly infested fields should be plowed and planted to a cultivated crop for a season." A color illustration of this species is given in Alexander (1932). Drawings of the seedling stages of A. gryposepala are given in Kummer (1951).

Agrimonia eupatoria was even used as a sleep aid, as the following Old English verse

testifies (Addison 1985):

If it be leyd under a mann's head He shall sleepyn as he were dead. He shall never drede ne wakyn, Till fro under his head it be taken.

Almost certainly any of our North American species would be as soporifically efficaceous.

Anti-ophidian properties were even ascribed to *A. eupatoria* (Law 1973). The plant was used in a charm to ward off snakes. One of its old English names was sticklewort, hence this bit of doggerel:

He that hath sticklewort by Knows no snake shall draw him nigh.

Agrimonia eupatoria was implicated by O'Donovan (1942) in the production of phytophotodermatitis in humans, but he established a probable connection between plant and patient in only one of 14 cases. Mitchell and Rook (1979) suggested that irritation (from the plant's trichomes?) rather than photosensitization was the cause of the condition. Later research showed that an alcoholic extract of A. eupatoria has "a very slight photosensitizing action" (Dijk 1963). Dijk and Berrens (1964) "assumed" but did not demonstrate that "Agrimonia contain[s] photosensitizing furocoumarins" as do other, much more potent photosensitizing plants, e.g., Ammi majus, Pastinaca sativa, and Ruta graveolens. Obviously more study of the problem is needed.

A tea can be made by steeping leaves and stems of A. parviflora and A. rostellata in boiling water, cooling, and serving with sugar or lemon (Cheatham and Johnston 1995). A similar tea, from A. eupatoria, is used in Britain as a "purifier of the blood" (Hulme 1912) and is said to be "particularly adapted to people who live poorly, and imperfectly digest their bad food" (Anonymous 1856).

Agrimonia eupatoria (leaves and stems) is a

minor dye plant, giving a stable yellow, gold, or orange depending on the mordant (Addison 1985; BBG 1984; Hutchinson 1972; S.M. Robertson 1973; Keville 1991; Lushchevskaya 1937; Usher 1974) and on the month of harvest (Johnson 1862). Our native species would probably be similarly useful.

Agrimonia eupatoria "contains tannin, and has been used in dressing leather" (Pratt 1905). Flowers of the species, with their "honey flavour," were once added to mead; the dried plant, with its fragrance, was included in "sweet sachets and pot-pourri" (Huxley 1992).

Several abnormalities have been noted in the development of flowers/inflorescences of Agrimonia, the reports all European (and thus the plant being probably A. eupatoria). Masters (1869) wrote that the genus is among those in which suppression of the androecium occurs most frequently and that leaves may develop in the center of a flower. Anomalies in the number of sepals, the position of bracts, and the number of bractlets are known (Phelouzat 1963). Fasciation in the inflorescence results in a compact, terminal mass of flowers, the whole being somewhat reminiscent of a head of a member of the Asteraceae (Phelouzat 1963; Schimper 1854). Fusion of two petals can form a structure similar to the keel of a legume flower (Moquin-Tandon 1842).

The petals of *A. eupatoria*, normally yellow, can on rare occasions, be white. Seeds of this white-flowered form, when planted, "bred true to white flower-colour" (Nelmes 1929).

Of the origin of the name Agrimonia we may choose from the four possibilities listed by Quattrocchi (2000): "Possibly from the Greek argemone, argemon, ancient name used by Dioscorides, Plinius and Galenus for the poppy; or from argemonion ancient Greek name applied by Dioscorides to the anemone; or from agros "field" and monos "alone, lonely"; or from agrios, agrimaios (agra) "wild." Another source (HcVN 2000) derived "agrimony" from a "Greek word describing plants which healed the eyes."

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## NOTE

Barn Owl (Tyto alba) Feeding Habits at Yellowbank Wildlife Management Area, Breckinridge County, Kentucky.—This study used pellet analysis to determine feeding habits of barn owls (Tyto alba) at the Yellowbank Wildlife Management Area (YWMA), Breckinridge County, Kentucky. We found that barn owl diets at YWMA consisted mainly of voles (Microtus spp.). These results are consistent with many past barn owl feeding habit studies.

Past studies suggest that barn owls consume a prevalence of small mammals (1, 2). However, larger animals (e.g., birds, reptiles, and amphibians), and insects (e.g., grasshoppers, beetles) are also often eaten (2, 3). Little is known, however, about barn owl feeding habits in the east-central U.S. We are unaware of any reports of barn owl feeding habits for Kentucky and Indiana, and there are only single reports for southern Illinois (4) and Ohio (5). The purpose of this study was to determine whether prey selection of barn owls in YWMA is consistent with other barn owl feeding habit studies.

Feeding habits of barn owls are generally examined through the dissection of regurgitated pellets. Barn owls consume their prey whole or largely so. Relatively weak digestive fluids secreted in the stomach dissolve the nutritious soft parts of the prey (1, 2). This is followed by the regurgitation of a tightly packed pellet consisting of indigestible fur, bones, insect exoskeleton, and/or feathers. Typically, two pellets are formed and cast each day, ca. 6.5 hr after ingestion of prey. One pellet is cast at a habitual roosting or nesting area, the other at night while the owl is foraging (6). Consumption of more than one meal during a 6.5-hr period may result in the casting of larger pellets containing remains of multiple meals (6). Thus, a record of past meals is created from undigested portions of prey (2, 7). The proportion of prey items captured by barn owls closely matches the proportion regurgitated, providing a relatively unbiased sampling of feeding habits (1, 2).

Pellets were gathered from beneath a nesting platform at YWMA during the 1998 nesting season. Two adult barn owls and seven young regurgitated the pellets.

Each dry pellet was weighed to the nearest 0.1 g, measured along the longest and shortest axes to the nearest 1 mm, soaked in water, then dissected using tweezers and dissecting probes. Skull and lower mandible remnants were counted and identified.

Small mammal capture records for YWMA and a list of mammal species occurring in Breckinridge County (both provided by the Kentucky Department of Fish and Wildlife) were used as a guide for preliminary identification. We used a reference skull collection (Mammal Collection, Southern Illinois University, Carbondale) to assist in species identification of bone fragments (8, 9, 10, 11, 12, 13).

We determined the percentage of each species within pellets by number and by mass, with estimates of mean mass (14, 15). Skull and mandible counts from large pel-

lets ( $\geq$  2.0 g) were compared to those of small pellets (< 2.0 g) with one-tailed t-tests adjusted for unequal sample size. Correlation analysis was used to assess the relationship between pellet mass and the number of prey items per pellet. Species proportions between small and large pellets were examined with chi-square, with a 2.0 g cutoff arbitrarily chosen after inspection of the distribution of pellet masses.

We examined 52 large and 151 small pellets for a total of 203 pellets. An average dry pellet weighed 1.6  $\pm$  0.1 g, and was 29.8  $\pm$  0.5 mm long  $\times$  20.9  $\pm$  0.4 mm wide. Small pellets contained fewer skulls (mean = 0.63) than large pellets (mean = 1.49);  $t_{\rm 67}$  =  $-6.79,\,P < 0.001$ ). Small pellets also had fewer mandibles (mean = 0.94) than large pellets (mean = 3.06;  $t_{\rm 66}$  =  $-7.80,\,P < 0.001$ ).

The most common genus among all pellets was Microtus (64%), primarily M. ochrogaster (n = 87), and M. pinetorum (n = 36; Table 1). Southern bog lemmings (Synaptomys cooperi; n = 15) were also prevalent. Species diversity increased as a function of pellet mass ( $r^2 = 0.24$ , P < 0.001). We were unable to classify remains from 29 of the 203 (14%) pellets. The proportion of unidentified specimens in small pellets was higher than in large pellets (27.9 vs. 3.8%, respectively;  $\chi^2 = 20.02$ , P < 0.001). The proportion of unidentified microtines in small pellets was nearly double that of large pellets (13% vs. 7.5%), though this difference was not significant, ( $\chi^2 = 1.60$ , P < 0.21). Composition of identified species was similar between small and large pellets, aside from a greater number of southern bog lemmings in the smaller pellets (13.0 vs. 2.0;  $\chi^2 = 6.35$ , P < 0.01). Pellets were rarely devoid of bones (n = 4). The only apparent difference in prey composition between pellet class sizes was in proportion of southern bog lemmings.

Barn owls are opportunistic foragers. Prevalence of prairie and woodland voles in the barn owl diet is likely a function of their relative ease of capture, concurrence of predator and prey activity periods, and prey abundance. Barn owls are most active at night and favor open country for foraging (3). Likewise, prairie and woodland voles remain on the ground (versus escaping up into the trees) when pursued. Woodland voles are also common in most stages of forest succession (12). Our data suggest that populations of shrews (Blarina sp.; Sorex spp.) and harvest mice (Reithrodontomys spp.) are not active during the same time period that barn owls hunt, do not forage in the same areas as barn owls, or use specific habitat components successfully to evade predation. In conclusion, our results indicate a preponderance of microtines in the diet of barn owls at YWMA and are consistent with other barn owl feeding habit studies (2, 4, 5, 16, 17).

We thank Dr. G. A. Feldhamer, and J. C. Whittaker, Department of Zoology, SIUC, and Dr. Alan Woolf, Cooperative Wildlife Research Laboratory and Department

Table 1. Numbers and percentages of known prey species found in 203 pellets from a pair of barn owls and their seven young at Yellowbank Wildlife Management Area, Breckinridge County, Kentucky.

Species (mean live mass [g])	Common name	Number	% by number	% by mass!
Microtus ochrogaster (42.5)	Prairie vole	87	50	61
Microtus pinetorum (32)	Woodland vole	36	21	19
Synaptomys cooperi (35.5)	Southern bog lemming	15	9	9
Blarina brevicauda (21.5)	Short-tailed shrew	9	5	3
Mus musculus (20.5)	House mouse	8	5 .	3
Cryptotis parva (5.3)	Least shrew	7	4	<1
Microtus pennsylvanicus (45)	Meadow vole	7	4	5
Reithrodontomys megalotis (13)	Harvest mouse	1	<1	<1
Unidentified bird <sup>2</sup>		4	2	NA
Total <sup>3</sup>		174		

For identified specimens only.

3 Excluding plant and insect contents.

of Zoology, SIUC, and numerous volunteers for laboratory assistance. \\ \\

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<sup>&</sup>lt;sup>2</sup> One specimen was likely a red-winged blackbird (Ageaius phoeniceus).

# Abstracts of Some Papers Presented at the 1999 Meeting of the Kentucky Academy of Science

#### AGRICULTURAL SCIENCES

Pesticide residue in soil and runoff: measurement and mitigation. GEORGE F. ANTONIOUS, Community Research Service, Atwood Research Facility, Department of Plant and Soil Science, Kentucky State University, Frankfort, KY 40601.

Soil erosion and runoff are some of the major means by which pesticides from agricultural fields enter streams, ponds, or lakes. The Water Quality Project at Kentucky State University (KSU) is evaluating best management practices for the growing of vegetable crops on highly erodible land (10% slope). Studies were conducted to determine the influence of landscape features on pesticide movement into runoff and infiltration water. Soil treatments (black plastic mulch and living fescue mulch) were used to reduce soil erosion and surface water runoff. Pesticides infiltration into the vadose zone were monitored using pressure-vacuum lysimeters (n = 27). A tipping bucket metering apparatus was used to collect runoff water following natural rainfall events. The impact of the soil mulches on movement of clomazone (a soil applied herbicide), dacthal (a pre-emergence non-systemic herbicide), and endosulfan (an insecticide) was measured under field conditions. Black plastic covers had no clear effect on reducing runoff volume or concentration of clomazone in runoff, while grass strips of 30-cm wide were very effective at reducing amounts of sediment in runoff. Plots planted with pepper intercropped with tomato as cover crop had 72% less runoff water and 79% less runoff sediment compared to plots planted with pepper only. Results indicated the vertical movement of clomazone, daethal, and endosulfan through the soil into the vadose zone. Cultivation of turf reduced runoff but did not reduce leaching of pesticides into the vadose zone. Our future objective at KSU is to study the potential of using soil amendments to improve soil quality, detoxify contaminants, and reduce erosion.

Identification of molecular markers that segregate in a simple Mendelian fashion in controlled crosses of pawpaw (Asimina triloba). SHAWN BROWN,\* TERA M. BONNEY, SNAKE C. JONES, and KIRK W. POMPER, Kentucky State University, Atwood Research Facility, Frankfort, KY 40601-2335.

The pawpaw (Asimina triloba) is a native plant found mainly in the southeastern and eastern United States. Its fruit has great potential as a new high-value crop in these regions. Although there are about 45 named pawpaw cultivars, breeding for improvement of desirable traits, such as improved fruit size and quality, is desirable. Our long-term goal is to utilize molecular marker systems to identify markers that can be used for germplasm diversity analysis and for the construction a molecular genetic map, where

markers are correlated with desirable pawpaw traits. The objective of this study was to identify random amplified polymorphic DNA (RAPD) markers that segregate in a simple Mendelian fashion in a controlled A. triloba cross. DNA was extracted from young leaves collected from field-planted parents and 20 progeny of the cross 1-7-1  $\times$  2-54, as well as 10 progeny of the reciprocal cross. The DNA extraction method used gave acceptable yields of about 7  $\mu g \, g^{-1}$  of leaf tissue. Additionally, sample 260/280 ratios were about 1.4, which indicated that the DNA was of high enough purity to be subjected to the RAPD methodology. Screening of 10-base oligonucleotide RAPD primers with template DNA from the parents and progeny of the cross is proceeding in an effort to identify RAPD markers that segregate in a simple Mendelian fashion.

The effect of transport density on survival of juvenile freshwater prawns (*Macrobrachium rosenbergii*). SHAWN COYLE, JAMES H. TIDWELL, AARON VANARN-UM,\* and CHARLES WEIBEL, Aquaculture Research Center, Kentucky State University, Frankfort, KY 40601.

In production of the freshwater prawn Macrobrachium rosenbergii, prawns (≥0.3 g) are nursed in indoor tanks then transported to ponds for growout. Stress during transport can produce immediate and undetected mortality after pond stocking. This study was designed to evaluate the effect of density on transport survival. Nine replicate styrofoam transport containers were prepared. Each contained a plastic bag with oxygen-saturated 22°C water with an atmosphere of 10-liter pure oxygen. Juvenile prawns weighing  $0.26 \pm 0.02$  g were randomly stocked into each of three replicate transport containers at 10, 25 or 50 g/liter of water, then sealed for 8 hours (maximum in-state transport period). At 8 hours post-stocking, bags were opened, water was sampled, and live and dead animals were separated and counted. Total ammonia-nitrogen and nitrite-nitrogen were significantly higher (P <0.05) in containers stocked at 50 g/liter than in containers stocked at either 10 or 25 g/liter, which were also significantly different (P < 0.05). Dissolved oxygen was significantly lower (P < 0.05) in transport containers stocked at 50 g/liter (1.3 mg/liter) than those stocked at 25 g/liter or 10 g/liter (1.6 mg/liter and 3.2 mg/liter, respectively), which were also significantly different (P < 0.05). Survival was significantly reduced (P < 0.05) in transport containers stocked at 50 g/liter (86.6%). Survival in containers stocked at 25 g/liter (93%) was significantly lower than containers stocked at 10 mg/liter (97.2%). These data indicate that transport densities greater than 10 g/liter should be avoided for transport ≥8 hours.

Suitability of the copepod Orthocyclops modestus as a live food for larval freshwater prawns, Macrobrachium ro-

senbergii. SHAWN COYLE,\* JAMES H. TIDWELL, AARON VANARNUM, and CHARLES WEIBEL, Aquaculture Research Center, Kentucky State University, Frankfort, KY 40601.

The cyclopoid copepod Orthocyclops modestus was evaluated for its suitability as a live food for larval freshwater prawns (Macrobrachium rosenbergii). Orthocyclops modestus was identified in preliminary screening as one of the few indigenous zooplanktors which tolerated the temperature (28-30°C) and salinity (10-14 ppt) conditions of larval prawn culture. To evaluate the suitability of these copepods as live food for larval prawns, mixed zooplankton were collected from a reservoir with a 250 µm zooplankton net. Zooplanktors were held at 10 ppt salinity for 24 hours to remove cladocerans and then screened (335 µm) to remove rotifer species leaving only copepods. The test system consisted of nine individual 250 ml rearing units in a recirculating system, with three replicates of each of three treatments. Treatment 1 contained only larval prawns (five 10-d old larvae, Stage 5.4 ± 0.9), Treatment 2 contained only copepods (185), and Treatment 3 a combination (5 larval prawns and 185 copepods). Densities for prawns were based on recommended prawn hatchery practices. Densities for copepods were based on recommended artemia feeding rates for Stage 5 prawn larvae. After 48 hours, prawn survival in Treatment 3 (87%) was significantly lower (P < 0.05) than in Treatment 1 (100%). Copepod survival was not significantly different between treatments (93.7%) indicating copepods were not consumed. Reduced prawn survival in Treatment 3 was likely due to high energy demands or physical trauma as prawn larvae attempted to capture active and wellarmored copepods. It appears that indigenous zooplankton show little promise as live foods in prawn hatchery production.

Use of hempseed meal, poultry by-product meal, and canola meal in practical diets without fish meal for sunshine bass. ANN M. MORGAN,\* CARL D. WEBSTER, KENNETH R. THOMPSON, and EBONY J. GRISBY, Aquaculture Research Center, Kentucky State University, Frankfort, KY 40601.

Sunshine bass (Morone chrysops  $\times$  M. saxatilis) is one cross of hybrid striped bass. Diets for sunshine bass use high percentages of fish meal (FM); however, FM is the most expensive ingredient in aquaculture diets. If FM can be replaced, diet costs may decrease. Four practical floating diets were formulated to contain 40% protein, similar energy levels, and without FM. A fifth diet was formulated to contain 30% FM and served as the control diet. Ten fish (21 g) were stocked into each of twenty 110-liter aquaria and were fed twice daily (07.30 and 16.00 hr) for 10 weeks. Diets were formulated to contain: Diet 1, sovbean meal (SBM) and meat-and-bone meal (MBM); Diet 2, SBM + MBM + hempseed meal (HSM); Diet 3, SBM and poultry by-product meal (PBM); and Diet 4, and SBM + MBM + canola meal (CM). At the conclusion of the feeding trial, percentage weight gain of fish fed Diet 1 was significantly (P < 0.05) higher (299%) compared to fish fed Diet 3 and Diet 4, but not different from fish fed Diet 2 and Diet 5. Percentage survival, amount of diet fed, and hepatosomatic index (HSI) of sunshine bass were not different (P > 0.05) among treatments and averaged 95%, 111 g of diet/fish, and 2.0% of body weight, respectively. Results from the present study indicate that diets without FM have potential for use in growing juvenile sunshine bass. Further research needs to be conducted on the diet formulations used in the present study and should be conducted in ponds.

The Kentucky State University pawpaw (Asimina triloba) project. KIRK W. POMPER,\* SNAKE C. JONES, EDDIE B. REED, and TERA M. BONNEY, Kentucky State University, Atwood Research Facility, Frankfort, KY 40601-2355.

Kentucky State University (KSU) has had a comprehensive pawpaw (Asimina triloba) research project since 1990, with the goal of developing the pawpaw into a new high-value tree fruit crop for limited resource farmers in Kentucky. An overview of recent developments at KSU concerning pawpaw variety trials, propagation, web site development, and germplasm collection and assessment, was presented. A pawpaw regional variety trial (RVT) was planted at KSU in 1998, consisting of 8 replicate trees of 28 standard pawpaw cultivars and advanced selections from the PawPaw Foundation breeding program. The RVT establishment rate, flowering, and growth data by variety was reported. The positive influence of shade on the growth and development of pawpaw seedlings in container production was discussed. A web site, http:// www.pawpaw.kysu.edu, has been developed and expanded for the dissemination of information on pawpaw to scientists, commercial growers and marketers. The National Clonal Germplasm Repository for Asimina spp. is located at KSU and hence, germplasm evaluation, preservation, and dissemination are a high priority for our program. In the spring of 1999, volunteers collected pawpaw leaf samples from 270 trees in 17 different states that will be used in molecular marker methodologies in order to assess genetic diversity across the pawpaw's native range. Pawpaw seedlings with promising fruit characteristics have been identified in our germplasm collection and have been propagated for further evaluation as potential cultivars for release by KSU.

Relative effectiveness of plant and animal source oils for control of air breathing insects. LEIGH ANNE VITATOE,\* AARON VANARNUM, SHAWN COYLE, and JAMES TIDWELL, Aquaculture Research Center, Kentucky State University, Frankfort, KY 40601.

Freshwater prawn juveniles (*Macrobrachium rosenbergii*) are stocked into ponds at extremely small sizes (0.2–0.5 g) and predation by air breathing insects can be a significant problem. The use of petroleum products to create a thin surface film and prevent insect respiration is an effective control but causes environmental concern. If

proven effective, plant- or animal-based oils may be safer. Menhaden fish oil (MO) and corn oil (CO) were evaluated at two application rates and compared to previously recommended petroleum product mixes for their ability to eliminate air-breathing insects. Petroleum product applications (controls) included 2:1 motor oil/diesel fuel combination (PCI) and 1:20 motor oil/diesel fuel combination (PCII) (previously recommended procedures). Glass aquaria with 0.107 m<sup>2</sup> surface area were used and filled with 6 liters of reservoir water. Each tank was stocked with five adult notonectids. Low rate applications were applied at 0.01 ml/m<sup>2</sup> and high rate applications at 0.03 ml/m<sup>2</sup>. There were three replications per treatment. Control tanks were stocked but not treated with oil. At the low rate the PCII and MO treatments both produced complete mortality by 2 hours post treatment while treatments PCI and CO did not result in complete mortality. At the high rate there was no significant difference (P > 0.05)among treatments in amount of time required to attain complete mortality. Menhaden fish oil appears to be an effective alternative to petroleum products for control of predaceous air breathing insects in larval shrimp and fish ponds. At a high rate of application corn oil may also be effective.

The effect of water temperature on the survival of adult freshwater prawns (*Macrobrachium rosenbergii*) held in tanks. CHARLES WEIBEL,\* JAMES H. TIDWELL, SHAWN COYLE, and AARON VANARNUM, Aquaculture Research Center, Kentucky State University, Frankfort, KY 40601.

Pond production of freshwater prawns (Macrobrachium rosenbergii) is becoming increasingly popular in Kentucky. As a seasonal crop, prawns must be harvested by mid-October to prevent losses. However, the highest demand for the product is during the holiday period of November through December. The ability to hold live freshwater prawns would allow producers to address this lucrative market. Temperature directly affects the metabolism of poikilothermic animals and reduced temperatures might increase survival under stressful conditions. Also, females carrying eggs at harvest are not considered desirable by some consumers and temperature could affect this trait. To address this, the effect of temperature on survival of adult freshwater prawns was evaluated under controlled conditions in tanks for 11 weeks. Freshwater prawns recently harvested from ponds (21.3 ± 1.7 g) were randomly stocked into nine 5700-liter tanks at 500 prawns/tank. There were three replicate tanks per temperature (17, 20, and 23°C). Prawns were fed a percentage of body weight at maintenance levels. After 76 days, average weight was not significantly different (P > 0.05) between treatments. Survival was significantly higher (P < 0.05) for animals held at 20° and 23°C (58 and 56%, respectively) than at 17°C (29%). The percentage of berried (egg carrying) females was significantly greater (P < 0.05) at 23° (13.2%) than at 20° or 17°C (0.2 and 0%, respectively). These data indicate that 20°C may be optimum for holding freshwater

prawns for market if berried females are undesirable. Holding temperatures near 17°C appear to represent stress conditions and result in high mortality.

#### **BOTANY & MICROBIOLOGY**

Inheritance of morphological and physiological characteristics in *Taraxacum officinale*. ANTON M. CLEM-MONS\* and DAVID L. ROBINSON, Department of Biology, Bellarmine College, Louisville, KY 40205.

Dandelion (Taraxacum officinale), an asexual species, produces achenes (seeds) apomictically. Three experiments were performed to explore the genetic variability and heritability of various morphological/ physiological traits that occur in natural populations. In the first study, achenes from a single dandelion population were subjected to a warm temperature treatment (37°C) for different time periods (0, 3, 4, 6, 8, 10, 14, 16 d), followed by a 4d incubation at 21°C, and a final cold treatment (5°C). Germination was assessed at the end of each temperature treatment. The experimental control (constant 21°C) exhibited the highest percent germination. In the other treatments, a positive correlation was found between the percent of ungerminated achenes and the duration of exposure to high temperature. To examine the heritability of achene heat tolerance, germinated achenes exposed to 8 d at 37°C, and then 21°C, were collected, grown to maturity, and allowed to flower and set seed. Achenes from these heat-tolerant plants (as well as control plants) were grown to maturity to evaluate the next generation. In the second study, achenes collected from dandelion populations occurring in five U.S. states were germinated and grown to maturity in a controlled environment. Analysis of leaf morphology revealed more variability between the five populations than within them. In the third experiment, achenes from 19 different fasciated (deformed) plants were germinated and grown in a controlled environment to examine the inheritance of their expressed fasciation. These experiments help to delineate the amount of genetic diversity in dandelion populations.

Biological effects of volatile emissions from cut turf. MARIA L. DAVIS\* and DAVID L. ROBINSON, Biology Department, Bellarmine College, Louisville, KY 40205.

Current research on the volatile emissions emanating from recently-mowed turfgrass indicates that these gases may have a significant impact on the environment. The primary purpose of this study was to observe the effect of gaseous emissions from different species on the rate of seed germination. In the first study, dandelion (Taraxacum officinale) and white snakeroot (Ageratina altissima [Eupatorium rugosum]) seeds were placed into sealable, plastic bags containing the freshly harvested foliage (cut into ca. 4 cm lengths) from a species of plant that commonly grows in turf. Seeds were treated with the emissions from one of seven plant species: Cynodon dactylon (L.) Pers., Lolium perenne, Leptochloa fascicularis, Taraxacum officinale, Trifolium repens, Glechoma hederacea, or Plantago lanceolata. Germination rates were examined over a 18-d

period, and the foliage-gas treatments that incurred the greatest effects were examined in more detail in a second experiment. In that study, A. altissima seeds were germinated in a replicated trial in the presence of cut foliage from one of four different species (L. perenne, T. officinale, T. repens, P. lanceolata) or an experimental control. No statistically significant differences were observed for rate of germination in any of the foliage treatments (including the control). In addition, the effect of these gases on crickets, a common grass-dwelling insect, will be discussed.

Identification of fecal coliform species from Lee's Branch at Midway, Kentucky. BEVERLY W. JUETT,\* DEBORAH EVEN, and GLENDA MARKER, Department of Biological Sciences, Midway College, Midway, KY 40347.

Coliform species were isolated and identified from 60 total coliform counts performed in Lee's Branch at Midway, KY. Water sampling was conducted six different times beginning in July 1996 and ending in September 1997. Total coliform counts were determined by the Standard Total Coliform Membrane Filter Procedure. Coliform species isolated from Endo agar plates were transferred to trypticase soy and MacConkey agar. The isolates were identified by conventional biochemical methods and Enterotube II. Identification of bacterial species in stream water provides basic knowledge of the microbial environment of freshwater and may serve as baseline data in determining if the fecal contamination of this stream is from animals or humans.

Continuation of a study on inheritance of achene characteristics in *Ageratina altissima*. JOANN M. LAU\* and DAVID L. ROBINSON, Department of Biology, Bellarmine College, Louisville, KY 40205.

Seed dormancy is a powerful means by which plants control when and where they occur. Three major sources for a population's variability for dormancy are genetic diversity, somatic polymorphism, and microsite/temporal/biological variability. The goal of this research was to explore the relative importance of these sources in regulating achene (seed) germination and dormancy in white snakeroot (Ageratina altissima [Eupatorium rugosum). Last year, white snakeroot achenes were selected for different germination and dormancy traits, grown to maturity and allowed to reproduce. The progeny (achenes) of these selections were then examined for different germination characteristics. Most (54%) of the progeny germinated before any cold treatment, whereas 12% germinated after a single cold treatment. Although, on average, there were no noticeable differences between the parental groups, there were obvious differences between individual selections. For instance, one plant (from a non-cold-requiring achene) produced achenes that exhibited 100% germination at room temperature, whereas another plant's progeny (selected for germination after 1 cold treatment) exhibited 32, 20 and 16% germination after 0, 1, and 2 cold

treatments, respectively. In another study, the effect of achene size on germinability was examined by partitioning achenes from a single population into four size categories. Even though the weight of the largest achenes was more than double the smallest there were no statistically significant differences in cumulative germination between any of the size categories. Therefore, if there is somatic polymorphism for achene dormancy in white snakeroot it may involve characteristics other than achene size.

### CELLULAR & MOLECULAR BIOLOGY

The GABAc rho1 and rho2 subunit genes are differentially expressed during pre- and postnatal development in the mouse. CHRISTY COLE,\* Transylvania University, Lexington, KY 40508; and MAUREEN McCALL, University of Louisville, Louisville, KY.

Most inhibitory neurotransmission in the central nervous system (CNS) is mediated either by glycine or gamma aminobutyric acid (GABA). GABA inhibition is mediated by one of three receptors: GABAa, GABAb, or GA-BAc. In the rodent, the GABAc receptor regulates the flow of chloride ions in the bipolar cells of the retina. The GABAc receptor may be made up of three types of subunits: rho1, rho2, and rho3. The GABAc rho2 subunit is predominant throughout the CNS, however, the GABAc rhol subunit seems to be predominant in only the bipolar cells of the retina. These subunits are expressed at different times in the development of the mouse. To determine at which point in the development of the mouse the GA-BAc rho1 and rho2 subunits are expressed, total RNA was extracted from mice at embryonic ages (E): E12.5, E14.5, E17.5 and from retinas of mice at postnatal ages (P): P1, P6, P13, P20. cDNA was reverse transcribed from all RNA samples and used in a PCR based strategy to determine the time course of the expression of both the GA-BAc rho1 and rho2 subunit genes. As a control for the amount of cDNA transcribed from each RNA sample, beta-actin was amplified from each cDNA sample. The expression of all three genes were tested individually using PCR, and the amplified products were visualized using electrophoresis. We have found that the GABAc rho1 gene and the rho2 gene are differentially expressed during pre- and postnatal development of the mouse. The GA-BAc rhol gene appears to begin to be expressed between P1 and P6. The GABAc rho2 expression is present in the youngest sample that we tested, E12.5. Although the GA-BAc rho1 and rho2 genes are thought to assemble into functional receptors in the adult, their differential expression during development suggests that the GABAc rho2 gene may play a different role in the developing embryo and may assemble homomeric receptors or associate with other GABA subunits to assemble functional receptors. These results will also aide in further research with transgenic and knockout mice which are being tested to determine the function of the GABAc receptor in retinal processing.

#### CHEMISTRY

Photoacoustic measurements in biological tissues and fluids. ANGELA L. NEWCOMB,\* Department of Science, Campbellsville University, Campbellsville, KY 42718; and JOEL MOBLEY and TUAN VO-DINH, Life Sciences Division, Oak Ridge National Laboratory, Oak Ridge, TN 37831.

The photoacoustic (PA) effect is the generation of acoustic waves by electromagnetic radiation. The magnitude of the PA response in a material is determined by its optical absorption. We have investigated the thermally-mediated PA response of mammalian brain tissue and glucose solutions to explore the utility of PA methods for biomedical sensing applications. We measured the PA spectra of tissues from the cerebrum of sheep between 500–700 nm using a tunable pulsed laser for excitation. We were able to differentiate between the gray and white matter in our samples, which could be useful in minimally invasive surgery. We were also able to detect the presence of glucose in solution, which may lead to a noninvasive method of measuring blood sugar.

### HEALTH SCIENCES

Analysis of a functional digital model of the mammalian basilar membrane. SHELLY FERRELL,\* MATTHEW E. KOGER, DAVID RICE, KENNETH M. MOORMAN, and PEGGY SHADDUCK PALOMBI, Transylvania University, Lexington, KY 40508.

The human auditory system functions as a processor of sound. The processing occurs in an analog setting where the sound stimulus is generated from a source and is directed toward the ear. From the pinna, the sound travels in the outer ear to the middle ear, through the cochlea where the basilar membrane vibrates to stimulate the hair cells. In this pathway the signal is transduced from a wave front to an electrical signal. This is also the critical point where the information is translated into a signal that the brain can process. The sound signal has a non-linear relation to the cochlear output. For a fully functioning computer model to simulate the process of basilar membrane function, it must contain the non-linear properties that the basilar membrane incorporates into the signal processing. Other minor inputs to the main sound wave are from sources such as pinna vibration, skull vibration, ossicular vibration, and residual tympanic membrane vibration. We attempted to evaluate a "best" computer model of auditory processing for accurate models that incorporate consistent non-linear properties and accurately use the other input conditions to fine tune and purify the simulation. We determined that the LUTEar model developed by Ray Meddis and his colleagues, contains a developed program that has moderately accurate non-linear properties. The consistency of the digital diagrams compared to physiological data is not as accurate as we would like. The overall processing shape is similar; however, the actual numbers are not within an acceptable percent error. Our next steps will be to reprogram portions of the models to achieve a

more accurate model, to correct minor non-linear property errors, and to increase incorporation of other stimuli to the processing output.

Examination of the validity of the pre-emphasis filter in an auditory system model. MATTHEW E. KOGER,\* SHELLY FERRELL, DAVID RICE, KENNETH M. MOORMAN, and PEGGY SHADDUCK PALOMBI, Transylvania University, Lexington, KY 40508.

LUTEar, a mammalian auditory system computer model, was examined to determine whether it could be manipulated to correctly model aged hearing. The validity of the pre-emphasis filter in LUTEar was investigated using previously published physiological data. The pre-emphasis filter corresponded with the outer and middle ear in the mammalian auditory system. It takes the input signal and passes it through a mathematical filter. This output is next sent to the basilar membrane filter. An investigation of the literature led to an understanding of the structure and function of the outer and middle ear; based on this understanding, physiological data was gathered from the literature and compared to data from LUTEar gathered by Meddis and Hewitt (1991). It was determined that LU-TEar amplified incoming sound in the outer and middle ear ten to twenty decibels less than what physiological data has found. However, it was determined that the basic filter shape of the amplification curve of the pre-emphasis filter closely matched the curve in physiological data. Next, the feasibility of altering the parameters of the preemphasis filter for aged hearing was explored. The preemphasis filter is a mathematical equation derived from a digital band-pass filter. The difficulty in altering this equation to accurately represent aged hearing was centered in the non-physiological based variables in the filter. It was determined that a greater study of this was needed. Therefore, changes that incorporate biological variables will have to be made to the digital band-pass filter in order to correctly model aged hearing.

A radioprotective drug combination in mice. K. S. KU-MAR, V. SRINIVASAN, D. L. PALAZZOLO,\* E. P. CLARK, and T. M. SEED, Radiation Medicine Department, Armed Forces Radiobiology Research Institute, Bethesda, MD 20889; and Pikeville College School of Osteopathic Medicine, Pikeville, KY 41501.

Thiol drugs are very effective radioprotective agents. However, the doses required to protect are also very toxic. One approach to minimize toxicity is to combine low doses of thiols with other non-thiol radioprotective agents. The thiol drug used was S-2-(3-methylaminopropylamino)-ethylphosphorothioic acid (WR-3689; 50 mg/kg), a methylated derivative of amifostine, which is used in chemotherapy to reduce the cis-platinum-induced nephrotoxicity. The non-thiols used were monophosphoryl lipid A (MPL, 0.5 mg/kg), an immunomodulator, and two prostaglandins—iloprost (ILO, 0.1 mg/kg), and misoprostol (MIS, 0.1 mg/kg). Individually, these doses of WR-3689 and MPL are known to be non-toxic, while ILO and

MISO are known to be toxic. In this study, male CD2F1 mice were given these agents intraperitoneally (IP) 30 minutes before irradiation with 10 Gy of 60Co at 1 Gy/minute, and survival was monitored for 30 d. When given individually, none of drugs were protective, as indicated by 0% survival. However, when all four agents were combined and given as a single IP injection, the survival rate increased to 50% and a dose reduction factor of 1.23 was calculated indicating significant radioprotection. In addition, the combined drug treatment appeared to be less toxic. These results indicate that the toxicity of radioprotectants at higher doses can be reduced by combining them at lower doses without compromising the radioprotective efficacy.

### MATHEMATICS

Superficial coset curiosities. JAMES B. BARKSDALE JR., Department of Mathematics, Western Kentucky University, Bowling Green, KY 42101.

This presentation exhibits several rudimentary propositions concerning coset notions from elementary group theory. Superficial curiosities which result from slight statement modifications of these fundamental theorems and relationships are then noted and discussed. Such presentation content could serve as an enrichment theme (or as a special project topic) for undergraduate mathematics courses.

Open mindedness to low-tech teaching methods. AUS-TIN FRENCH, Department of Mathematics/Physics/ CSC, Georgetown College, Georgetown, KY 40324.

An effective teaching method involving (1) no note-taking by students (but the students have a clear set of notes), (2) no homework grading by the instructor for outside work, (3) a text that costs a maximum of \$9, (4) a system where it is extremely hard for a student to cheat and where the student's grade measures what the student knows (not that the student was in a group with someone that knew something and they got that student's grade), (5) a mixture of practice and creativity is expected by the student, (6) knowing absolutely perfectly some hard problem types is rewarded, (7) where two overhead projectors are used, and (8) transparencies are made from neat pencil-written notes . . . will be introduced. The concept of overheads as the text will be shown. Surgical strike class questions, microquizzes, presentations, and jugular problems will be described in this unique grading system, which is simply fun to teach by and does not squeeze all of the blood out of the teacher's turnip to use this method. Two overheads are used in conjunction with transparencies from pencil-written overheads comprising the text for the course. This talk should help you to inform the technologically gullible that just because something involves high-tech methods, it does not necessarily mean it is best to use technology and that all need to be open-minded about other teaching methods.

### PHYSICS & ASTRONOMY

Searching for hydrogen gas in shell galaxies. SEPPO LAINE,\* Department of Physics and Astronomy, University of Kentucky, Lexington, KY 40506; STEPHEN T. GOTTESMAN and KARL E. HAISCH JR., Department of Astronomy, University of Florida, Gainesville, FL 32611; and BENJAMIN K. MALPHRUS, Department of Physical Sciences, Morehead State University, Morehead, KY 40351.

Recent images of the hydrogen line emission from shell galaxies have given new clues about the origin of the shells. Detection of gaseous material associated with these optical features gives us an opportunity to investigate the formation mechanism of shells and even of the associated lenticular or elliptical galaxies. Therefore, we sought to expand the sample of shell galaxies that could be mapped in the 21-cm hydrogen line. We used the 140-ft telescope of the National Radio Astronomy Observatory at Green Bank, WV to attempt to detect the 21-cm emission line in 9 shell galaxies. The observations were centered at the frequency of the spectral line, corresponding to the systemic velocity of the underlying galaxy. The total onsource integration times varied between 3 and 11 hours. We detected emission from the direction of two shell systems, NGC 3610 and NGC 4382. However, NGC 4382 has a nearby disk galaxy within the 20 arcmin beam of the telescope. Therefore, the more likely candidate for detection of gas associated with shells is in NGC 3610. Since the signal extends over a rather narrow range of velocities and the optical images show a small, possibly interacting galaxy within the radio telescope beam, the explanation for this detection is unclear. Recent Very Large Array 21cm observations of NGC 3610 should resolve the source of the hydrogen line emission.

#### PHYSIOLOGY & BIOCHEMISTRY

Effect of Tamoxifen, Genistein, and vitamin E on the activity of the cysteine proteases Cathepsin L and Cathepsin B and their endogenous inhibitors in human androgen-independent prostate cancer cell lines. T. BURC-CHIO,\* E. HUGO, M. MARKEY, B. PHILLIPS, A. BURNS, D. DARIA, J. THOMPSON, T. HOLDEN, E. McDONOUGH, G. HENSON, D. FRITZ, B. HURST, and J. H. CARTER, Wood Hudson Cancer Research Laboratory, Newport, KY 41071.

Metastatic prostate cancer is a leading cause of death in men. Initially, most forms of this cancer are repressed by the removal of androgens and several treatment courses are based on this phenomenon. Growth inhibition by androgen removal, however, is often transient, with the carcinoma growth becoming independent of exogenous androgens. The proliferation of tumor cells correlates with the levels of the lysosomal proteases Cathepsin B (CB) and Cathepsin L (CL) as well as the endogenous inhibitors (CPI) of these enzymes. The role of these proteins in the progression of prostate carcinoma (CaP) is most likely one of protein turnover, however, we cannot rule out the pos-

sibility of some interaction with the process of metastasis. We have previously demonstrated that Tamoxifen ((Z)2-[4-(1,2-diphenyl-1-butenyl) phenoxy]-N, N-dimethylethanamine 2-hydroxy-1,2,3- propanetricarboxylate) (Tam), an antiestrogen, Genistein (4',5,7-trihydroxyisoflavone) (Gen), a soy phytoestrogen, and vitamin E ( $\alpha$ -tocopherol succinate) (VitE) have a potent growth inhibitory effect on three cell lines derived from metastatic prostate adenocarinomas: LNCaP, DU-145, and PC-3. In this study, we have examined the effects of these compounds on the levels of CB, CL and CPI both intracellularly and secreted into the medium. We find that after exposure to these compounds the levels of CB and CL drop below the level of detection. CPI levels, while falling dramatically, remain detectible and are found to have the same specific activity (% protease inhibition/mg protein) as in growing cells.

Examination of the stability of the acidic domain of Narginine dibasic convertase, an opioid specific peptidase. KELLI CARPENTER\* and EVA CSUHAI, Department of Chemistry, Transylvania University, 300 North Broadway, Lexington, KY 40508.

N-arginine dibasic convertase (NRDc) is a metallopeptidase. NRDc has been cloned and sequenced and has been shown to contain an unusually large number of acidic residues. It has been suggested that polyamines may regulate the activity of NRDc by binding to acidic residues located at the anionic domain of the enzyme [Csuhai et al. (1998) Biochemistry 37, 3787-3794]. The acidic domain of NRDc was produced and purified as a GST fusion protein in these experiments in order to run stability tests in the future by placing the fusion protein in a variety of media. The GST fusion proteins containing the acidic domain of mouse NRDc or human NRDc, as well as a control containing GST alone were scanned in a Circular Dichroism Spectrophotometer with and without the polyamine spermine in order to determine any structural effects spermine may have on the enzyme. There was no significant change in the percentage of alpha-helix, betaturn, beta-sheet, or random portions in the enzyme. Therefore, spermine does not appear to have any significant effect on the secondary structure of the fragment containing the acidic domain of NRDc. Mouse NRDc was scanned in a Circular Dichroism Spectrophotometer with a temperature gradient in order to determine the effect that spermine has on the enzyme's heat stability. A concentration of 1 mM of spermine was used in the tests. There was no distinct heat transition observed under these conditions. Therefore, based on these results, spermine does not appear to affect the structure and stability of the acidic domain of NRDc.

### SCIENCE EDUCATION

Classroom cheating: A survey. JOHN G. SHIBER, Prestonsburg Community College, Prestonsburg, KY 41653.

A survey of 877 high school students from four eastern Kentucky counties was conducted to determine the stu-

dents' attitude toward classroom cheating. Results: 38% said cheating in school is alright; 62% said it is wrong but 82% have done it; 95% have witnessed it; 67% would let friends copy their test answers if asked; 81% agreed plagiarism is cheating, and 56% admitted having done it, although 34% didn't know at the time they were plagiarizing; 33% do not regard copying friends' homework as cheating; 65% believe teachers cheat when they give grades higher or lower than students earn; 41% have been given a higher grade than they deserved at one time or other but only half brought it to the teacher's attention; and 47% have noticed teachers overlooking cheating, and, of these, 41% attribute it to teachers not caring what students do, 35% to favoritism, and 24% to their not wanting to embarrass the student or their desire to help the student pass the course. Compared to responses of 630 community college students to the same survey questions (Shiber 1999), those reported here suggest that high schoolers have a far more liberal attitude towards cheating and much greater experience with it than their older counterparts. The two groups' ideas about why students cheat are similar, however, with laziness to do work ranking first, lack of study time due to extra-curricular activities or family/job obligations second, too much pressure on good grades third, ineffective test-monitoring fourth, and, finally, many said students cheat because everybody cheats.

Megalitter: An Appalachian deformity. J. SMITH\* and J. SHIBER, Prestonsburg Community College, Prestonsburg, KY 41653.

Three designated points, located on or near the banks of eastern Kentucky's Paintsville Lake, were systematically investigated for the first time for the presence, type, and abundance of megalitter over a 2-month period. Megalitter was collected by hand from each site every 2 weeks in March/ April 1999. It was sorted according to the type of material it was made from, counted, and recorded. By far, the most abundant type of megalitter was plastic, then styrofoam, paper/clothing, metals, and glass. As the weather warmed and more people frequented the park, the occurrence of megalitter tripled at the three locations. The incidence of plastic more than quadrupled. This pilot study indicates that a severe megalitter problem continues to exist within the environs of Paintsville Lake Park, despite the annual community clean-ups and daily efforts of the park personnel. The seriousness of illegal dumping and littering, not only in this park, but all over eastern Kentucky and other regions of the state, as witnessed by volunteers in the government-funded P.R.I.D.E. Program, makes it imperative that a mandatory educational program on waste management be implemented by the Commonwealth in the earliest stages of public schooling and maintained within the science curriculum throughout high school. Furthermore, appropriate penalties for littering and dumping can only be truly effective if more manpower is provided to enforce them, both in our public recreation facilities and the outlying communities.

#### ZOOLOGY & ENTOMOLOGY

The effects of pH levels on diversity and density of aquatic microorganisms. CHRIS ALTMAN and POLLY FROSTMAN,\* Department of Biology, Transylvania University, Lexington, KY 40508.

To look at the potential effects of acid rain, we designed an experiment to determine whether or not there is a significant correlation between pH levels and diversity and/ or density of aquatic microorganisms. Water was collected from a man-made lake and divided into nine equal samples. Each sample was examined to determine density and diversity under normal pH conditions. Then 20 ml of each sample (1-9) were removed. Half of these samples had their pH lowered to 6-6.5 and the other half had their pH lowered to 4 by adding aqueous sulfuric acid (H2SO4). Then the density and diversity of microorganisms present in each acidified sample was examined. Each set of data showed significant differences between the control samples and the acidified samples suggesting that the pH levels do affect the diversity and density of aquatic microorganisms. The pH levels used in this experiment are comparable to levels that could be caused by acid rain suggesting that acid rain can overcome the buffering capacity of water and affect aquatic ecosystems.

Optimal foraging in chickens (*Gallus domesticus*). SU-MEET R. BHATT and BRIAN A. CAUDILL,\* Department of Biology, Transylvania University, Lexington, KY, 40508.

We conducted a series of experiments to determine whether domesticated chickens (Gallus domesticus) were able to forage optimally under three different feeding conditions. Our study was conducted on three barnyard roosters and 10 hens fed at two feeding stations 3 m apart. In our first experiment, the chickens were presented with three times as much food at one feeding station as at the other. In the second experiment, chickens were fed twice as fast at one station as at the other. And in the third experiment, whole corn kernels were fed at one station, and an equivalent number of half com kernels were fed at the other. The chickens showed a significant preference for feeding at the high quantity feeding station (first experiment) and the high feeding rate station (second experiment), but showed no significant preference for large food items over small (third experiment). This suggests that chickens are capable of making some basic decisions about where to forage to maximize their energy intake per unit time, although they did not seem very capable at discriminating between different sizes of food items that were presented in equal quantities.

Effects of continual food restriction on reproductive development and body organs in male house mice (*Mus musculus*). MICHAEL B. BOONE\* and TERRY L. DERTING, Department of Biological Sciences, Murray State University, Murray, KY 42071.

Prior research has shown that moderate levels of intermittent food restriction have no negative effect on the reproductive development of male house mice. We determined the effects of moderate but continual food restriction on de-

velopment of the reproductive and digestive systems in postweaning male house mice. We tested the null hypothesis that continual food restriction does not affect the level of testosterone or the masses of reproductive and body organs. Food intake of post-weaning males was restricted daily in a manner that prevented growth in overall body mass of the male. The males were sacrificed after 21 d and their level of testosterone and the wet and dry masses of their reproductive and other body organs were measured and compared with similar data from control males. The wet and dry masses of the testes were significantly lower in the food-restricted males. In contrast, testosterone levels of the food-restricted males were 55% higher, but not significantly different, compared with control males. The wet and dry mass of the stomach, but not the cecum, small intestine, and colon, was significantly heavier in food-restricted males compared with control males. Our results, in combination with those of studies using intermittent food-restriction, indicate that when food resources are limited energy is allocated preferentially to processes necessary for reproduction (e.g., testosterone production for spermatogenesis). A negative impact of food restriction on reproductive processes in mammals may only occur when food restriction is severe.

Richness of terrestrial vertebrate species in Kentucky. MATTHEW L. COLE,\* TERRY L. DERTING, and HOWARD WHITEMAN, Department of Biological Sciences, Murray State University, Murray, KY 40271.

To explore the diversity of terrestrial vertebrate species in Kentucky, we examined the distribution of species using richness (number of species) as an index of diversity. We evaluated species richness for amphibians, breeding birds, mammals, reptiles, and all terrestrial vertebrates combined using two different regional delineations from the US Forest Service: ecoregions and physiographic provinces. Using species ranges obtained from the scientific literature, species richness was calculated for each pixel ( $600 \times 600 \text{ m}$ ) across the state. We overlaid the species richness maps with coverages of the ecoregions and provinces of Kentucky using a geographic information system (CIS). Among the ecoregions and provinces, mean species richness differed most for reptiles, particularly among the squamates. Mean species richness was highest for reptiles in the Mississippi Embayment and lowest in the Cumberland Mountains, a difference of 78%. In contrast, mean species richness was highest for amphibians and mammals in the Cumberland Mountains and lowest in the Bluegrass Region, a difference of approximately 20%. The variation in mean species richness for breeding birds was minimal, differing by no more than 10% among the physiographic provinces. Largely due to the high number of reptile species, mean species richness was 10-15% higher in the westernmost ecoregion and provinces compared with other geographic areas. Areas of highest species richness of terrestrial vertebrates were the Mississippi Embayment, Mississippi Alluvial Basin, and Cumberland Mountains. Management plans that protect these "hotspots" of species diversity will be necessary if the biodiversity of vertebrates in Kentucky is to be maintained.

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### **INDEX TO VOLUME 61**

### Compiled by Varley Wiedeman

A. nipponica x A. pilosa var. japoni-

A. parviflora, 146-148, 150, 151-

A. pubescens, 146, 147, 150, 151-

A. rostellata, 146, 147, 150, 151-153

Air breathing insects, control of,

A. striata, 146, 155, 157, 158

ca. 157

A. pilosa, 157, 158

153, 157

A. suaveolens, 146

Agropyron spicatum, 92

Aimophila aestivalis, 127

A. sylvatica, 146

Abies fraseri, 52 Abietinella abietina, 117 ABSTRACTS FROM 1999 KAS MEETING, 165-172 Acadian, 87 Accipiter striatus, 127 Acentrella ampla, 20 Acer pensylvanicum, 54 A. rubrum, 11, 54 A. saccharum, 11, 54 A. spicatum, 117 Achalarus lyciades, 86 Acipenser fulvescens, 125 Acleris youngana, 106 Aconitum uncinatum, 117 Acornshell, 130 Acorus calamus, 23, 26, 27 Acrobasis vaccinii, 106 Acroneuria, 14, 15 A. caroliniensis, 20 Actinastrum gracillimum, 36, 37 Actitis macularia, 127 Adams cave beetle, lesser, 125 ADAMS, KELLY, 62, 99 Adiantum capillus-veneris, 117 Adlumia fungosa, 117 Admiral red. 87 white, 87 Aeschnidae, 21 Aesculus octandra, 11 A. pavia, 117 Agalinis auriculata, 117 A. obtusifolia, 117 A. skinneriana, 117 Agalis milberti, 87 Agastache scrophulariifolia, 117 Ageaius phoeniceus, 164 Ageratina altissima, 167, 168 inheritance of achene characteristics in, 168 A. luciae-brauniae, 117 Agraulis vanillae, 87 Agricultural sciences, 165-167 Agrimonia, 146-162 in Kentucky, 146-162

A. eupatoria, 144, 147, 154, 155,

A. eupatoria × A. procera, 157A. gryposepala, 117, 146, 147, 150,

A. microcarpa, 146, 154, 155, 157

151-154, 157, 158

157-159

A. incisa, 156

166-167 Alabama lip fern, 118 Alabama shad, 125 Alasmidonta marginata, 124 A. atropurpurea, 124 Algae, of Land Between the Lakes, 34-45 Allegheny chinkapin, 118 Allegheny stonecrop, 122 Alligator gar, 125 Alligator snapping turtle, 127 Allocapnia sp., 20 Alloperla sp., 20 Alosa alabamae, 125 ALTMAN, CHRIS, 171 Amblyopsis spelaea, 125 Amblyscirtes aesculapius, 86 A. belli, 86 A. hegon, 86 A. vialis, 86 Amelanchier laevis, 54 Ameletidae, 20 Ameletus, 15 A. sp., 20 American barberry, 117 American bison, 130 American bittern, 127 American brook trout, 126 American burying beetle, 125 American chaffseed, 122 American chestnut, 52, 118 American coot, 127 American copper, 87 American cow-wheat, 120 American crow, 53, 54 American crow-wheat, 120 American frog's-bit, 120 American golden-saxifrage, 118 American goldfinch, 56 174

American lady, 87 American lily-of-the-valley, 118 American redstart, 56 American snout butterfly, 87 American speedwell, 123 American water-pennywort, 119 American wintergreen, 121 Amianthium muscitoxicum, 117 Ammi majus, 159 Ammocrypta clara, 125 A. vivax, 130 Ammodramus henslowii, 127 Amphibians, 126-127 Amphinemura, 15 A. delosa, 20 Amphipod, 125 Amphipod, Bousfield's, 124 Amphiuma, three-toed, 126 Amphiuma tridactylum, 126 Amsonia tabernaemontana var. gattingeri, 117 Anaea andria, 87 Anartia jatrophae, 87 Anas clypeata, 127 A. discors, 127 Anatrytone logan, 86 Ancylid, domed, 123 Ancyloxipha numitor, 86 Anemone, Canada, 117 Anemone canadensis, 117 Angelica, filmy, 117 Angelica triquinata, 117 Angled riffleshell, 130 Anglepod, Carolina, 120 Anguispira rugoderma, 123 Anhinga, 130 Anhinga anhinga, 130 Animal source oils, 166-167 effectiveness on air breathing insects, 166-167 Animals, 123-128, 130 Ankistrodesmus falcatus, 34, 35 A. spiralis, 36, 37 Anodontoides denigratus, 124 Anomodon rugelii, 117 Antaeotricha osseela, 105 Anthocharis midea, 87 Anthomyia, 155 ANTONIOUS, GEORGE F., 23, 165 Antroselatus spiralis, 123 Apalone mutica mutica, 127 Apamea, undescribed species, 107

Aphrodite fritillary, 87	A. pilosus var. priceae, 117	Bay starvine, 122
Apidae, 155	A. pratensis, 117	Beak-rush, woodland, 122
Apioblasma haysiana, 130	A. radula, 117	Beaked-rush
Apios priceana, 117	A. saxicastellii, 117	globe, 121
Apis, 155	Asterocampa celtis, 87	tall, 121
Aporrectodea, 3	A. clyton, 87	Bean
Appalachian blue, 87	Atalopedes campestris, 86	Cumberland, 124
Appalachian brown, 87	Atlides halesus, 87	rayed, 124
		Bear, black, 128
Appalachian bugbane, 118	Atractosteus spatula, 125	Bearded skeleton, 119
Appalachian grizzled skipper, 125	Atrichapogon sp., 21	Beaver cave beetle, 125
Appalachian rosinweed, 122	Atrytonopsis hianna, 86	BEBE, F. N., 108 Beebalm, spotted, 120
Appalachian sandwort, 120	Auditory system model, 169	Beetle
Appalachian sedge, 118	pre-emphasis filter in, 169	American burying, 125
Appalachina chilhoweensis, 123	Aureolaria patula, 117	Ashcamp cave, 125
Aquatic microorganisms, 171	Autochton cellus, 86	beaver cave, 125
density, 171	Azalea, hoary, 121	bold cave, 125
diversity, 171	Azure	cave, 125
effects of pH levels on, 171	dusky, 87	Clifton cave, 125
Arabis hirsuta var. adpressipilis, 117	spring, 87	concealed cave, 125
A. missouriensis, 117	Azygiidae, 60–62, 99–104	Cub Run Cave, 125
A. perstellata, 117		Garman's cave, 125
Archilochus colubris, 53	Baby-blue-eyes, small-flower, 121	Greater Adams Cave, 125
Ardea alba, 127	Bachman's sparrow, 127	hidden cave, 125
A. herodias, 127	Bachman's warbler, 130	icebox cave, 125
Aristida ramosissima, 117	Baetidae, 20	lesser Adams cave, 125
Armoracia lacustris, 117	Baetis sp., 20	limestone cave, 125
Armored rocksnail, 123	B. flavistriga, 20	Louisville cave, 125
Arrow head, 27	B. intercalaris, 15, 20	Old Well Cave, 125
Arrow-wood, Missouri, 123	B. tricaudatus, 15, 20	Roger's cave, 125 round-headed cave, 125
Arrowhead, delta, 122	BAIRD, NANCY DISHER, 83	scholarly cave, 12
grass-leaf, 122	Bald eagle, 127 Baltimore checkerspot, 87	sixbanded longhorn, 125
	Bambusina brebissonii, 44, 45	Stevens Creek Cave, 125
sessile-fruit, 122	Banded darter, 70	surprising cave, 125
Arrowwood, downy, 123	Banded 87	Tatum Cave, Little Black Moun-
Arthrodesmus convergens, 40, 41	Bank swallow, 128	tain, 50–59
A. extensus, 40, 41	Baptisia australis var. minor, 117	Bird-voiced treefrog, 126
A. octocornis, 40, 41	B. bracteata var. leucophaea, 117	Birds, breeding, 127–128, 130
Artificial nest density, 46–49	B. tinctoria, 117	Bishop's-weed
effect on Canada Goose, 46–49	Barbara's-buttons, 120	eastern mock, 121
Ascia, 155	Barbed rattlesnake-root, 121	mock, 121
Asellidae, 22	Barberry, American, 117	Nuttall's mock, 121
Ashcamp cave beetle, 127	Barbicambarus cornatus, 124	Bison, American, 130
Ashy darter, 70, 126	Barking treetrog, 127	Bittern
Asimina triloba, 163, 164	BARKSDALE, JAMES B., JR., 170	American, 127
Asio flammeus, 127	Barn owl, 128	least, 127 Bivalva, 20
A. otus, 127	feeding habits, 163–164	Black bear, 128
Aster	Barrens silky aster, 117	Black buffalo, 126
barrens silky, 117	Bartonia virginica, 117 Bartramia longicauda, 127	Black locust, 54, 56
eastern silvery, 117	Bashful bulrush, 122	Black lordithon rove beetle, 125
low rough, 117	Bass	Black swallowtail, 87
Rockcastle, 117	hybrid striped, 166	Black tern, 130
Tennessee, 117	sunshine, 166	Black-and-white warbler, 56
Texas, 117	diets for, 166	Black-crowned night-heron, 127
white heath, 117	Basswood, 54	Black-throated blue warbler, 54–56
	Bat	Blackberry, smooth, 122
whorled, 117	evening, 128	Blackbird, red-winged, 164
Aster acuminatus, 117	Rafinesque's big-eared, 128	Blackburnian warbler, 50, 53, 55,
A. concolor, 117	Virginia big-eared, 128	127
A. drummondii var. texanus, 117	Battus philenor, 86	Blackfin sucker, 126
A. hemisphericus, 117	B. polydamas, 87	Blackfoot quillwort, 120

journai o.	t the Kentucky Reademy of Sex
Blackside dace, 126	Bombus, 155
Blacktail redhorse, 126	Bonasa umbellus, 53
Blacktail shiner, 125	BONNEY, TERA M., 165, 166
Bladderpod	BOONE, MICHAEL B., 171
Lescur's, 120	Borer moth, rattlesnake-master, 125
Lesquereux's, 120	Bos bison, 130
-1 11	Botany & Microbiology, 167–168
Bladderwort, greater, 123 Bladetooth, Virginia, 123	Botaurus lentiginosus, 127
1	Botrychium matricariifolium, 117
Blarina sp., 163 B. brevicaudad, 164	B. oneidense, 117
-1 1 1 100	Botryococcus braunii, 36, 37
Blazingstar, slender, 120	
Bleuter, 124 Bloodfin darter, 70	Bottlebrush crayfish, 124 Bottomland lichen, 117
Blossom	Bottomland lichen, 117 Bousfield's amphipod, 124
tubercled, 130	
	Boyleinia aconitifolia 118
yellow, 130	Boykinia aconititolia, 118
Blotched chub, 126	Brachythecium populeum, 117 Branched three-awn 117
Blue grees drooping 121	Branta canadensis, 46
Blue grass, drooping, 121 Blue heron	
	Braun's rock-cress, 117 Breeding birds, 127–128, 130
great, 127	- 1
little, 127	Brighteye darter, 126 Brilla sp., 22
Blue jasmine leather-flower, 118	
Blue jay, 54	Bristly sedge, 118  Broad banded water spake, 127
Blue monkshood, 117	Broad-banded water snake, 127 Broad-leaf golden-aster, 119
Blue mud-plantain, 117	
Blue scorpion-weed, 121	Broad-winged skipper, 86 Broadleaf water-milfoil, 121
Blue water I., 23, 26, 27	Broadwing sedge, 118
Blue wild indigo, 117 Blue	Broken-dash
	northern, 86
Appalachian, 87 eastern tailed, 87	southern, 86
	5
marine, 87	Brook Jamprey
silvery, 87 Blue-flower coyote-thistle, 119	Brook lamprey mountain, 126
Blue-headed vireo, 55	northern, 126
PO 3	southern, 126
Blue-joint reed 118 Blue-star, eastern, 117	Brook saxifrage, 118
Blue-winged teal, 127	Brook snaketail, 125
m1 11 11 00	Brook trout, American, 126
Blue-winged warbler, 55 Bluebreast darter, 70	Broomrape, Louisiana, 121
Bluecurls, narrow-leaved, 123	Brown bog sedge, 118
Bluegill, 73	Brown creeper, 127
Bluets	Brown elfin, 87
clustered, 121	Brown madtom, 126
Michaux's, 119	Brown, Appalachian, 87
Blunt mountain-mint, 121	BROWN, D. KEVIN, 133
Blunt-lobe grapefern, 117	BROWN, HETTI A., 164
Bluntface shiner, 125	BROWN, SHAWN, 165
Bobolink, 127	Brown-headed cowbird, 50, 54, 56
Bog club-moss	Bryocamptus morrisoni elegans, 124
northern, 120	Bryum cyclophyllum, 117
southern, 120	B. miniatum, 117
Bog goldenrod, southern, 122	Bubulcus ibis, 127
Bog lemming, southern, 163, 164	Buckeye
Bog rush, 120	common, 87
Bog sedge	red, 117
brown, 118	Buckley's goldenrod, 122
prickly, 118	Buffalo clover, 123
BOIADGIEVA, EMILIA 62, 99	running, 123
Bold cave beetle, 125	Buffalo, black, 126
Boloria bellona, 87	Bugbane, Appalachian, 118
B. selene myrina, 87	Bulbochaete varians, 36, 38
Boluteloua curtipendula, 118	Bull paspalum, 121
	Laskan,

Bulrush bashful, 122 Hall's, 122 river, 122 slender, 122 softstem, 23, 26, 27 Bunchflower, Virginia, 120 Bunting, indigo, 56 Bur-reed, large, 122 Burbot, 126 Burhead, 119 dwarf, 119 Burnet, Canada, 122 BURNS, A., 170 BURRCCHIO, T., 170 Burrowing mayfly, 125 robust pentagenian, 130 Burying beetle, American, 125 Bush's muhly, 120 **Bush-clover** round-head, 120 tall, 120 Butler's quillwort, 120 Buttercup, 27 Butterflies, Kentucky, 86-87 Butterfly, American snout, 87 Button, wrinkled, 123 Cabbage white, 87

Cabomba caroliniana, 118 Caddisflies, 15 Helma's net-spinning, 125 limnephilid, 125 Cadmium, effect of on rats, 108-114 Caecidotea sp., 22 C. barri, 124 Cajun dwarf crayfish, 124 Calamagrostis canadensis var. macouniana, 118 C. porteri ssp. insperata, 118 C. porteri ssp. porteri, 118 Calephelis borealis, 87 C. mutica, 87 Calla lily, 27 Callirhoe alcaeoides, 118 Callophrys augustuius, 87 C. grynea, 87 C. henrici, 87 C. irus, 87 C. niphon, 87 Calopogon tuberosus, 118 Calopterygidae, 21 Calopteryx sp., 21 Caloptilia fraxinella, 105 Caltha palustris var. palustris, 130 Calycanthus floridus var. glaucus, Calycopis cecrops, 87 Calylophus serrulatus, 118 Cambarellus puer, 124 C. shufeldtii, 124 Cambaridae, 22

Cambarus parvoculus, 124

C. veteranus, 124 Catharus fuscescens, 53, 55 Chestnut, American, 52, 118 CAMPBELL, JULIAN J. N., 88 Catspaw, 124 Chestnut-sided warbler, 54, 55, 56 Campephilus principalis, 130 white, 130 Cheumatopsyche sp., 21 Campostoma anomalum, 75 Cattails, 23-27 C. helma, 125 Canada anemone, 117 Cattle egret, 127 Chickens, optimal foraging in, 171 Canada burnet, 122 CAUDILL, BRIAN A., 171 Chinkapin, Allegheny, 118 Canada frostweed, 119 CAUDILL, TERESA L., 46 Chionodes, undescribed species, Canada goose, 46-49 Cave beetle, 125 105 effect of artificial nest density on, Ashcamp, 125 C. adamas, 106 beaver, 125 C. aruns, 106 effect of wetland size on, 46-49 bold, 125 C. baro, 106 in constructed wetlands, 46-49 Clifton, 125 C. hapsus, 105 Canada warbler, 50, 53, 55, 128 concealed, 125 C. sevir, 106 Garman's, 125 C. suasor, 105 Canadian yew, 123 Chlidonias niger, 130 Canby's mountain-lover, 121 hidden, 125 Canis lupus, 130 icebox, 125 Chironomidae, 22 C. rufus, 130 lesser Adams, 125 Chloralictus, 155 Canola meal, 166 limestone, 125 Chlorococcales, 34 in diets for fish, 166 Louisville, 125 Chlorogonium euchlorum, 34, 35 Capniidae, 20 Roger's, 125 Chloroperlidae, 20 round-headed, 125 Caprifoliaceae, 30-33 Chlorophyta, 34-45 Cardinal flower, 27 scholarly, 125 Chondestes grammacus, 127 Cardinal, northern, 53, 56 surprising, 125 Chrysemys picta dorsalis, 127 Cardinalis cardinalis, 53 Cave isopod, Clifton, 124 Chrysogonum virginianum, 118 Carduelis tristis, 53, 56 Cavefish Chrysosplenium americanum, 118 Carex aestivalis, 118 northern, 125 Chub C. alata, 118 southern, 126 blotched, 126 C. appalachica, 118 Cavesnail, shaggy, 123 flame, 130 C. atlantica ssp. capillacea, 118 Ceanothus herbaceus, 118 flathead, 126 C. austrocaroliniana, 118 Cedar sedge, 118 gravel, 130 C. buxbaumii, 118 Celastrina argiolus ladon, 87 hornyhead, 126 C. comosa, 118 Celastrina ebenina, 87 sicklefin, 126 C. crawei, 118 sturgeon, 126 C. neglectamajor, 87 C. crebriflora, 118 Celephelis mutica, 125 Chubsucker, lake, 126 C. decomposita, 118 Celithemis verna, 125 Cimicifuga rubifolia, 118 C. gigantea, 118 Cellular & Molecular Biology, 168 Cinygmula, 15 C. hystericina, 118 Central mudminnow, 126 C. subaequalis, 20 C. joorii, 118 Central stoneroller, 75 Circaea alpina, 118 Centroptilum sp., 20 C. juniperorum, 118 Circus cyaneus, 127 C. lanuginosa, 118 Ceratopogonidae, 21 Cirriphyllum piliferum, 117 C. leptonervia, 118 Ceratopsyche sparna, 21 Cistothorus platensis, 127 C. reniformis, 118 CLARK, E. P., 169 Cercyonis peagala, 87 C. roanensis, 118 Certhia americana, 127 Classroom cheating, 171 C. rugosperma, 118 Cerulean warbler, 50, 53 Cleft phlox, 121 Cervus elaphus, 130 C. seorsa, 118 starry, 121 C. stipata var. maxima, 118 Chaetophorales, 36 Clematis crispa, 118 CLEMMONS, ANTON M., 167 Chaffseed, American, 122 C. straminea, 118 C. tetanica, 118 Clethrionomys gapperi maurus, 128 Chain pickerel, 126 Carolina anglepod, 120 Chalosyne gorgone, 87 Clifton cave beetle, 125 Carolina fanwort, 118 Channel darter, 70 Clifton cave isopod, 124 Carolina larkspur, 119 Charadrius melodus, 116 Clifty covert, 123 Carolina parakeet, 130 Charidryas nycteis, 87 Climbing fumitory, 117 Clioperla clio, 20 Carolina satyr, 87 Chat, yellow-breasted, 53 Cloak, mourning, 87 Carolina wren, 56 Checkered white, 87 Carolina yellow-eye, 123 Clonophis kirtlandii, 127 Checkerspot Closteriopsis longissima, 36, 37 CARPENTER, KELLI, 171 Baltimore, 87 Carya spp., 11, 54 Gorgone, 87 Closterium abruptum, 36, 39 C. ehrenbergii, 36, 39 Carya aquatica, 118 silvery, 87 C. setaceum, 36, 39 Castanea dentata, 52, 118 Cheilanthes alabamensis, 118 Clouded skipper, 86 C. feei, 118 C. pumila, 118 Clouded sulphur, 87 Castilleja coccinea, 118 Chelone obliqua Cloudless sulphur, 87 Catchfly var. obliqua, 118 ovate, 122 var. speciosa, 118 Cloudywing royal, 122 Chestnut lamprey, 126 confused, 86

110	Journal
1 00	
northern, 86	
southern, 86	
Clover	
buffalo, 123	
running buffalo, 123	
Club-moss	
northern bog, 120	
southern bog, 120	
Clubshell, 124	
Tennessee, 124	
Clubtail, elusive, 125	
Clustered bluets, 121	
Clustered poppy-mallow,	118
Crustered poppy-manow,	110
Coachwhip, eastern, 130	
Coal skink	
northern, 127	
southern, 127	
Coastal Plain sedge, 118	
Cobweb skipper, 86	
Coccyzus americanus, 53	20 05
Coelastrum cambricum,	36, 37
C. microporum, 36, 37	
Coeloglossum viride var.	virescens,
118	
Coil, punctate, 123	
Colombos curetus 52	
Colaptes auratus, 53	
COLE, CHRISTY, 168	
COLE, MATTHEW L.,	
Coleochaete orbicularis,	36, 38
C. scutata, 36, 38	
Coleoptera, 21	
Colias cesonia, 87	
C. eurytheme, 87	
C. philodice, 87	
Collinsonia verticillata, 13	18
Columbine duskywing, 86	3
COMBS, MICHAEL S.,	
Combshell	
Cumberlandian, 124	
round, 130	
Comma	
buckeye, 87	
checkered skipper, 86	
eastern, 87	
gray, 125	
green, 87, 125	
green, 67, 125	
gray, 87	
Common moorhen, 127	
Common raven, 127	
Common roadside skippe	er, 86
Common silverbell, 119	
Common sootywing, 86	
Common wood-nymph, 8	277
Common yellowthroat, 5	
Compassplant, 122	
Compton tortoise shell, 8	37
Comptonia peregrina, 11	8
Concealed cave beetle, 1	
Conchapelopia sp., 22	
Coneflower, sweet, 122	
Confused cloudywing, 86	1
Conjurer's-nut, 121	
Conradina verticillata, 11	8
Constempellina sp., 22	
1	

,
Constructed wetlands, 23–29
Canada Goose in, 46–49
Contopus virens, 53
Conuropsis carolinensis, 130
Convallaria montana, 118
Coot, American, 127
Copepod, 124
Copper iris, 120
Copper
American, 87
bronze, 87
Copperbelly water snake, 127
Coral, 87
Corallorhiza maculata, 118
Cordulegaster, 14
C. sp., 21
Cordulegasteridae, 21
Coreopsis pubescens, 118
Cormorant, double-crested, 128
Corn snake, 127
Corvus brachyrhynchos, 53
C. corax, 127
C. ossifragus, 127
Corydalidae, 21
Corydalis, pale 118
Corydalis sempervirens, 118
Corynoneura sp., 22
Corynorhinus rafinesquii, 128
C. townesendii virginianus, 128
Cosmarium baileyi, 39, 40
C. bipunctatum, 39, 40
C. biretum, 39, 40
C. blyttii, 39, 40
C. botrytis, 39, 40
C. depressum, 39, 40
C. granatum, 39, 40
C. margaritatum, 39, 40
C. meneghinii, 40, 41
C. moniliforme, 40, 41
C. monmorme, 40, 41
C. nymannianum, 40, 41
C. obtusatum, 40, 41
C. orthostichum, 40, 41
C. ovale, 40, 41
C. phaseolus, 40, 41
C. porrectum, 40, 41
C. portienum 40, 41
C. portianum, 40, 41
C. pyramidatum, 40, 41
C. subtumidum, 40, 41
C. turpinii, 40, 41
Cosmocladium pusillum, 40, 41
Cotton mouse, 128
Cotton- tawny, 119
COVELL, CHARLES V., JR., 86
105
Covert, cliffty, 123
Cow-parsnip, 119
Cow-wheat, American, 120
Cowbird, brown-headed, 50, 54, 56
COYLE, SHAWN, 165, 166, 167
Coyote-thistle, blue-flower, 119
Craba cuneifolia, 119
Crabapple, southern, 120
Cracking pearlymussel, 130
Crambidae, 105, 106

Crawe's sedge, 118 Crawfish frog, northern, 127 Crayfish, 124, 125 bottlebrush, 124 Cajun dwarf, 124 Crittenden, 124 dwarf, 124 Louisville, 124 Cream wild indigo, 117 Creek heelsplitter, 124 Creekshell Kentucky, 124 mountain, 124 Creeper, brown, 127 Creeping St. John's-wort, 119 Creole pearly-eye, 87 Crescent pearl, 87 tawny, 87, 125 Cress glade, 120 lake, 117 necklace glade, 120 Cricotopus trifascia, 22 Crinkled hair 119 Crittenden crayfish, 124 Cross-leaf milkwort, 121 Crossline skipper, 86 American, 53, 54 fish, 127 Crow-wheat, American, 120 Crucigenia tetrapedia, 36, 37 Crustacea, 22 Crustaceans, 124-125 Cryocopus pileatus, 53 Cryptobranchus allaganiensis alleganiensis, 126 Cryptotis parva, 164 Crystal darter, 130 Crystallaria asprella, 130 CSUHAI, EVA, 171 Cub Run Cave beetle, 125 Cuckoo, yellow-billed, 53 Cumberland bean, 124 Cumberland elktoe, 124 Cumberland leafshell, 130 Cumberland papershell, 124 Cumberland rosemary, 118 Cumberland sandwort, 120 Cumberlandia monodonta, 124 Cumberlandian combshell, 124 Cupped vertigo, 124 Curtis' goldenrod, 122 Cut turf, 167-168 volatile emissions from, 167-168 Cutleaf meadow-parsnip, 123 Cutleaf water-milfoil, 121 Cyanocitta cristata, 53, 54 Cyllopsis gemma, 87 Cymophyllus fraserianus, 118 Cynodon dactylon, 167 Cyperus plukenetii, 118

Crater, queen, 123

o 711 u 110	70	72 1
Cyperus, Plukenet's, 118	D. tigrina, 53	Duskytail darter, 67–76, 126
Cypress darter, 126	D. virens, 53	Duskywing
Cypress minnow, 126	Dero nivea, 20	columbine, 86
Cypress-swamp sedge, 118 Cyprinella camura, 125	DERTING, TERRY L., 171	dreamy, 86 funereal, 86
C. venusta, 125	Deschampsia cespitosa ssp. glauca, 119	Horace's, 86
Cypripedium candidum, 118	D. flexuosa, 119	Juvenal's, 86
C. kentuckiense, 118	Desmidium aptogonum, 44	mottled, 86
C. parviflorum, 119	D. grevillii, 44	sleepy, 86
C. reginae, 119	D. swartzii, 44, 45	wild indigo, 86
Cyprogenia stegaria, 124	Dewberry, Wharton's, 122	zarucco, 86
71 8 8 7	Diamesa sp., 22	Dusted skipper, 86
Dace	Diana fritillary, 87	Dwarf burhead, 119
blackside, 126	Diasemiodes nigralis, 106	Dwarf crayfish, 124
longnose, 126	Dichanthelium boreale, 119	Cajun, 124
Dainty sulphur, 87	Dicranodontium asperulum, 117	Dwarf dandelion, western, 120
Dalea purpurea, 119	Dicranota sp., 22	Dwarf sundew, 119
Danaus gilippus, 87	Dictyosphaerium pulchellum, 36, 37	
D. plexippus, 87	Didiplis diandra, 119	Eagle, bald, 127
Dandelion, 167	Digenea, 99–104	Eallophrys irus, 125
western dwarf, 120	Digenetic trematode, 60-63	Earleaf false foxglove, 117
DARIA, D., 170	Digital model	Early 87, 125
Dark-eyed junco, 50, 53, 55, 127	analysis of, 169	Earthworms, 1–5
Darter	of mammalian basilar membrane,	Eastern blue-star, 117
ashy, 70, 126	169	Eastern coachwhip, 130
banded, 70	DILLARD, GARY E., 34	Eastern comma, 87
bloodfin, 70	Dion skipper, 86	Eastern eulophus, 121
bluebreast, 70	Diplectrona, 10, 13, 15	Eastern hellbender, 126
brighteye, 126	D. modesta, 21	Eastern mock bishop's-weed, 121
channel, 70	Diplocladius sp., 22	Eastern phoebe, 53
crystal, 130	Diploperla robusta, 20	Eastern pine elfin, 87
cypress, 126	Diptera, 21, 155	Eastern puma, 130
duskytail, 67–76, 126	Disporum maculatum, 119	Eastern ribbon snake, 127
emerald, 70	Dixa sp., 22	Eastern silvery aster, 117
firebelly, 126	Dixidae, 22	Eastern slender glass lizard, 127
goldstripe, 126	Docidium baculum, 36, 39	Eastern small-footed myotis, 128
greenside, 70	D. undulatum, 36, 39	Eastern spotted skunk, 128
gulf, 126	Dodecatheon frenchii, 119	Eastern tailed blue, 87
johnny, 126	Dogface, southern, 87	Eastern turkeybeard, 123
least, 130	Dolichonyx oryzivorus, 127	Echinodorus berteroi, 119
longhead, 126	Dollar sunfish, 126	E. parvulus, 119
olive, 126	Domed ancylid, 123	Eclipidrilus sp., 20
rainbow, 70	Double-crested cormorant, 128	Ectopistes migratorius, 130
relict, 126	Double-ringed pennant, 125	Ectopria, 15
scaly sand, 130	Dove, mourning, 53	E. nervosa, 21
Shawnee, 126	Downy arrowwood, 123	Edward's, 87
smallscale, 126	Downy goldenrod, 122	Eel-grass, 123
speckled, 70	Dragon-head, slender, 130	Eggert's sunflower, 119
spotted, 126	Dreamy duskywing, 86	Eggleston's violet, 123
swamp, 126	Dromedary pearlymussel, 130	Egret
tippecanoe, 70	Dromus dromas, 130	cattle, 127
western sand, 125	Dropping blue 121	great, 127
Dayle Maria I 167	Dropseed northern, 122	Egretta carulea, 127 EISENHOUR, DAVID J., 67
DAVIS, MARIA L., 167	rough, 122	Eiseniella, 3
Decapoda, 22	Drosera brevifolia, 119	Elanoides forficatus forficatus, 130
Delaware skipper, 86	D. intermedia, 119	Elaphe guttata guttata, 127
Delicate vertigo, 124 Delphinium carolinianum, 119	Dryobius sexnotatus, 125	Elaphria cornutinis, 107
Delta arrowhead, 122	Dryopidae, 21	E. festivoides, 107
Dendroica caerulescens, 53, 54	Dryopteris carthusiana, 119	Eleocharis olivacea, 119
D. cerulea, 50, 53	D. ludoviciana, 119	Elfin
D. coronata, 53	Duke's skipper, 86, 125	brown, 87
D. fusca, 50, 53, 127	Dun skipper, 86	eastern pine, 87
D. pensylvanica, 53, 54	Dusky azure, 87	frosted, 87
r,,,,,	,	· · · · · · · · · · · · · · · · · · ·

Henry's, 87 Elimia ebenum, 60-63 E. semicarinata, 60, 61 Elk. 130 Elktoe, 124 Cumberland, 124 Elm, September, 123 Elmidae, 21 Elodea nuttallii, 119 Elusive clubtail, 125 Elymus species, 88–98 E. elymoides, 88 E. glabriflorus, 88, 89, 92 E. glaucus, 88, 92, 93 ssp. glaucus, 96 ssp. jepsonii, 93, 96 ssp. mackenzii comb nov., 88-98 ssp. virescens, 93 var. minor, 88 E. macgregorii sp. nov., 88-98 E. mackenzii, 93 E. stebbinsii, 92 E. svensonii, 119 E. trachycaulus, 93, 95 E. villosus, 92 E. virginicus, 88, 89, 92 var. glabriflorus, 92 var. intermedius, 89, 92 var. jejunus, 88, 89, 92 var. minor, 92 Emerald darter, 70 Emperor hackberry, 87 tawny, 87 Empididae, 22 Empidonax minimus, 127 E. virescens, 53 Enchanter's-nightshade, small, 118 Endopiza yaracana, 106 Enodia anthedon, 87 E. creola, 87 E. portlandia missarkae, 87 Entodon brevisetus, 117 Epargyreus clarus, 86 Epeorus, 10, 15 E. prob. namatus, 20 Ephemera guttulata, 20 E. simulans, 20 Ephemerella, 10, 15 E. inconstans, 125 E. prob. auravilii, 20 Ephemerellid mayfly, 125 Ephemerellidae, 20 Ephemeridae, 20 Ephemeroptera, 20 Epioblasma arcaeformis, 130 E. biemarginata, 130 E. brevidens, 124 E. capsaeformis, 124 E. flexuosa, 130 E. florentina florentina, 130 E. florentina walkeri, 130 E. lewisii, 130

E. obliquata obliquata, 124

E. obliquata perobliqua, 130 E. personata, 130 E. propinqua, 130 E. sampsonii, 130 E. stewardsonii, 130 E. torulosa rangiana, 124 E. torulosa torulosa, 130 E. triquetra, 124 Epiphytic sedge, 118 Epling's hedge-nettle, 122 Eremosphaera viridis, 34, 35 Erimystax insignis, 126 Erimystax x-punctatus, 130 Erimyzon sucetta, 126 Eriophorum virginicum, 119 Erora laetus, 87, 125 Eryngium intergrifolium, 119 Erynnis baptisiae, 86 Erynnis brizo, 86 E. funeralis, 86 E. horatius, 86 E. icelus, 86 E. juvenalis, 86 E. lucilius, 86 E. martialis, 86 E. zarucco, 86 Erythronium rostratum, 119 Esox lucius, 75 E. niger, 126 Etheostoma spp., 60 E. baileyi, 70 E. blennioides, 70 E. caeruleum, 70 E. camurum, 70 E. chienense, 126 E. cinereum, 70, 126 E. flabellare, 67, 68 E. fusiforme, 126 E. lynceum, 126 E. maculatum, 126 E. microlepidum, 126 E. microperca, 130 E. nigrum susanae, 126 E. parvipinne, 126 E. percnurum, 67–76, 126 E. proeliare, 126 E. pyrrhogaster, 126 E. sanguifluum, 70 E. stigmaeum, 70 E. swaini, 126 E. tecumsehi, 126 E. tippecanoe, 70 E. virgatum, 60, 61 E. zonale, 70 Euastrum abruptum, 40, 43 E. affine, 40, 43 E. ansatum, 40, 43 E. binale, 40, 43 E. denticulatum, 40, 43 E. didelta, 40, 43 E. elegans, 40, 43 E. evolutum, 40, 43

E. insulare, 40, 43

E. verrucosum, 40, 43 Euchloe olympia, 87 Eufala skipper, 86 Eukiefferiella spp., 22 Eulophus, eastern, 121 Eumeces anthracinus anthracinus, 127 E. anthracinus pluvialis, 127 E. inexpectatus, 127 Eupatorium maculatum, 119 E. rugosum, 167 E. semiserratum, 119 E. steelei, 119 Euphorbia mercurialina, 119 Euphydryas phaeton, 87 Euphyes dion, 86 E. dukesi, 86, 125 E. vestris, 86 Euptoieta claudia, 87 Eurema lisa, 87 E. nicippe, 87 European skipper, 86 Eurycea guttolineata, 126 Eurylophella funeralis, 20 Eurytides marcellus, 87 EVÉN, DEBORAH, 168 Evening bat, 128 Evening primrose, 121 yellow, 118 stemless, 121 Everes comyntas, 87 Extirpated biota of Kentucky, 115-Fagus grandifolia, 11 Falcate Orange Tip, 87 Falco peregrinus, 127 Falcon, peregrine, 127 False foxglove earleaf, 117 pale, 117 spreading, 117 ten-lobe, 117 False gromwell hairy, 121 soft, 121 western, 121 False hellebore, 120 small-flowered, 120 False mallow, hispid, 120 False solomon-seal, starry, 120 Fameflower limestone, 122 roundleaf, 123 Fanshell, 124

Fanwort, Carolina, 118

Fecal coliform species, 168

identification of, 168

Feniseca tarquinius, 87

from Lee's Branch, 168

Fat pocketbook, 124

Fèe's lip fern, 118

Farancia abacura reinwardtii, 127

Alabama lin 110	Europe hit American 100	
Alabama lip, 118	Frog's-bit, American, 120	Glyceria acutiflora, 119
Fèe's lip, 118 southern maidenhair, 117	Frog northern crawfish, 127	Glyph
southern shield wood, 119	northern leopard, 127	Maryland, 123
spinulose wood, 119	Frosted elfin, 87, 125	sculpted, 123
FERRELL, SHELLY, 169	FROSTMAN, POLLY, 171	Glyphyalinia raderi, 123
Fescue, 56	Frostweed	G. rhoadsi, 123
Festuca arundinacea, 56	Canada, 119	Gnaphalium helleri var. micraden-
F. elatior, 27	plains, 119	ium, 119
Fetterbush, 120	Fulica americana, 127	Goatweed leafwing, 87 Goera sp., 21
Few-flowered scurf-pea, 121	Fumitory, climbing, 117	Goeridae, 21
Fiddleleaf	Fumonelix wetherbyi, 123	Goerita, 15
one-flower, 119	Fundulus chrysotus, 126	G. betteni, 21
ovate, 119	F. dispar, 126	Gold-banded skipper, 86
Fiery skipper, 86	Funereal duskywing, 86	Golden topminnow, 126
Filmy angelica, 117	Fusconaia subrotunda subrotunda,	Golden-aster, broad-leaf, 119
Fimbristylis, hairy, 119	124	Golden-crowned kinglet, 55
Fimbristylis purberula, 119		Golden-saxifrage, American, 118
Finely-nerved sedge, 118	GABAc rho1 subunit genes, 168	Golden-star, 119
Fir, Fraser, 52	in the mouse, 168	Golden-winged warbler, 50, 53, 55,
Firebelly darter, 126	GABAc rho2 subunit genes, 168	128
Fish crow, 127	in the mouse, 168	Goldenclub, 121
Fishes, 125–126, 130	Gallinula chloropus, 127	Goldenrod
Five-lined skink, southeastern, 127	Gallus domesticus, 171	Buckley's, 122
Flame chub, 130	Gammarus bousfieldi, 124	Curtis', 122
Flathead chub, 126	Gar, alligator, 125	downy, 122
Floater, green, 124	Garman's cave beetle, 125	Rand's, 122
Fluted kidneyshell, 124	Gastropods, 123–124	Roan Mountain, 122
Fly-poison, 117	Gattinger's lobelia, 120	Short's, 122
Flycatcher, least, 127	Gaywings, 121	southern bog, 122
Forcipomyia sp., 21	Gelechiidae, 105	squarrose, 122
Forestiera ligustrina, 119	Gemmed satyr, 87	white-haired, 122
Forkshell, 130	Genistein, 170–171	Goldfinch, American, 56
Fox grape, northern, 123	effect on Cathepsin B, 170–171	Goldstripe darter, 126
Foxglove	effect on Cathepsin L, 170–171	Golenkinia radiata, 36, 37
earleaf false, 117	effect on cysteine proteases, 170–	Gomphidae, 21
pale false, 117	171	Gonatozygon brebissonii, 36, 39
spreading false, 117 ten-lobe false, 117	Gentian	Gorgone checkerspot, 87
Fraser fir, 52	prairie, 119	GOTTESMAN, STEPHEN T., 170
Fraser's loosestrife, 120	showy, 119	Gracillariidae, 105
Fraser's sedge, 118	yellow, 119	Grama, side-oats, 118
Fraxinus americana, 54	Gentiana decora, 119	Grape honeysuckle, 120
French's shooting-star, 119	G. flavida, 119	Grape northern fox, 123
FRENCH, AUSTIN, 170	G. puberulenta, 119 Geometrid moth, 125	sand, 123
Freshwater mussels, 124, 130	Geometridae, 105, 107	Grapefern
Freshwater prawns, 165, 166	Geothlypis trichas, 53	blunt-lobe, 117
copepods as live food for, 165-	Gian hyssop, purple, 117	matricary, 117
166	Giant swallowtail, 87	Grapeskin, glassy, 124
effect of water temperature on	GIBSON, LORAN D., 105	Grass
survival, 167	Gillmeria pallidactyla, 107	bearded skeleton, 119
transport density on survival, 165	Glade cress, 120	blue-joint reed, 118
Fringed nut-rush, 122	necklace, 120	branched three-awn, 117
Fringeless orchid, white, 121	Glandularia canadensis, 119	crinkled hair, 119
Fritillary	Glass lizard, eastern slender, 127	drooping blue, 121
Aphrodite, 87	Glassy grapeskin, 124	hair, 120
Diana, 87	Glassywing, little, 86	June, 120
great-spangled, 87	Glaucopsyche lygdamus, 87	northern witch, 119
gulf, 87	Glechoma hederacea, 167	pale manna, 123
meadow, 87	Gleditsia aquatica, 119	Porter's reed, 118
regal, 87, 125	Globe beaked-rush, 121	purple sand, 123
silver-bordered, 87	Gloeocystis planctonica, 34, 35	reed bent, 118
variegated, 87	Glossosoma sp., 21	reed canary, 23, 26, 27
FRITZ, D., 170	Glossosomatidae, 21	sharp-scaled manna, 119

journal	Talle Trendenty Trendently of Select	100 01(2)
shortleaf skeleton, 119	juniper, 87	Heptageniidae, 20
tufted hair, 119	northern, 87, 125	Heracleum lanatum, 119
Grass-leaf arrowhead, 122	red-banded, 87	Hermeuptychia sosybius, 87
Grass-of-parnassus	striped, 87	HERNER-THOGMARTIN, JEN-
kidney-leaf, 121	white-m, 87	NIFER H., 163
largeleaf, 121	Hairy false gromwell, 121	Heron
Grass-pink, 118	Hairy fimbristylis, 119	great blue, 127
Grassleaf mud-plantain, 119	Hairy groovebur, tall, 117	little blue, 127
Gratiola pilosa, 119	Hairy hawkweed, 119	Herzogiella turfacea, 117
G. viscidula, 119	Hairy ludwigia, 120	Hesperia leonardus, 86
Gravel chub, 130	Hairy rock-cress, 117	H. metea, 86
Gray comma, 87, 125	Hairy skullcap, 122	H. sassacus, 86
Gray hairstreak, 87	Hairy snout-bean, 121	Heteranthera dubia, 119
Gray myotis, 128	HAISCH, KARL E., JR., 170	H. limosa, 119
Gray treefrog, 127	Halesia tetraptera, 119	Heterotheca subaxillaris var. latifol-
Gray wolf, 130	Haliaeetus leucocephalus, 127	ia, 119
Great blue heron, 127	Halictidae, 155	Hexastylis contracta, 119
Great egret, 127	Halictus, 155	H. heterophylla, 119
Great Plains ladies'-tresses, 122	Hall's bulrush, 122	Hexatoma sp., 22
Great purple, 87	Haploperla sp., 20	Hickory, 87
Great-spangled fritillary, 87	Haplotaxida, 20	water, 118
Greater Adams Cave beetle, 125	Harelip sucker, 130	Hidden cave beetle, 125
Greater bladderwort, 123	Harrier, northern, 127	Hieracium longipilum, 119
Greater prairie-chicken, 130 Greater redhorse, 130	Harvest mouse, 163, 164	Hispid false mallow, 120
Grebe, pied-billed, 128	Harvester, 87	Hoary azalea, 121
Green comma, 87, 125	Hawk, sharp-shinned, 127	Hoary mode groups 121
Green floater, 124	Hawkweed, hairy, 119 Hayhurst's scallopwing, 86	Hoary nock orange, 121
Green orchis, long-bract, 118	Health Sciences, 169–170	Hoary-pea, spiked, 123 Hobomok skipper, 86
Green treefrog, 127	Heartleaf plantain, 121	HOLDEN, T., 170
Green water snake, Mississippi, 127	Heartleaf	Homoeosoma deceptorium, 107
Green-and-gold, 118	southern, 119	Honeysuckle
Greenside darter, 70	variable-leaved, 119	grape, 120
GRISBY, EBONY J., 166	Heath aster, white, 117	wild, 120
Grizzled skipper, 86	HEDEEN, STANLEY E., 6	Hooded merganser, 127
Appalachian, 125	Hedeoma hispidum, 119	Hooded warbler, 54–56
Gromwell	Hedge-hyssop	Horace's duskywing, 86
hairy false, 121	shaggy, 119	Hornsnail
soft false, 121	Short's, 119	rugged, 123
western false, 121	Hedge-nettle, Epling's, 122	shortspire, 123
Groovebur, tall hairy, 117	Heelsplitter, creek, 124	Hornyhead chub, 126
Grosbeak, rose-breasted, 50, 53, 55,	Helianthemum bicknellii, 119	Horse-balm, whorled, 118
56, 128	H. canadense, 119	House mouse, 164
Ground juniper, 120	Helianthus eggertii, 119	body organs, 171
Gulf darter, 126	H. silphioides, 119	effect of food restriction, 171
Gulf fritillary, 87	Helichus fastigiatus, 21	male, 171
Gymnopogon ambiguus, 119	Helicodiscus notius specus, 123	reproductive development, 171
G. brevifolius, 119	H. puntatellus, 123	Houstonia serpyllifolia, 119
XX 11 07	Hellbender, eastern, 126	HUGO, E., 170
Hackberry emperor, 87	Hellebore	Hummingbird, ruby-throated, 53
Hair 120	false, 120 small-flowered false, 120	HURST, B., 170
Hair	Helma's net-spinning caddisfly, 125	Hyalotheca dissiliens, 44
crinkled, 119	Helmitheros vermivorus, 53	H. mucosa, 44 Hybognathus hayi, 126
tufted, 119	Helvibotys pseudohelvialis, 105	H. placitus, 126
Haircap moss, 117 Hairstreak	Hemerodromia sp., 22	Hybopsis amnis, 126
Acadian, 87	Hemistena lata, 130	Hybrid striped bass, 166
banded, 87	Hemitremia flammea, 130	Hydrobaenus sp., 22
coral, 87	Hempseed meal, 166	Hydrocotyle americana, 119
early, 87, 125	in diets for fish, 166	Hydrogen gas in shell galaxies, 170
Edward's, 87	Henry's elfin, 87	Hydrolea ovata, 119
gray, 87	Henslow's sparrow, 127	H. uniflora, 119
great purple, 87	HENSON, G., 170	Hydrophyllum virginianum, 119
hickory, 87	Heptageniid mayfly, 125	Hydropsyche betteni, 21
•		

Hydropsychidae, 21
Hyla avivoca, 126
H. cinerea, 127
H. gratiosa, 127
H. versicolor, 127
Hylephila phyleus, 86
Hylocichla mustelina, 53
H. mustelina, 56
Hymenoptera, 155
Hypericum adpressum, 119
H. crux-andreae, 119
H. nudiflorum, 120
H. pseudomaculatum, 120
Hyssop, purple giant, 117

Icebox cave beetle, 125 Ichthyomyzon castaneus, 126 I. fossor, 126 I. gagei, 126 I. greeleyi, 126 Icteria virens, 53 Icterus spurius, 53 Ictinia mississippiensis, 127 Ictiobus niger, 126 Illinois pondweed, 121 Immyria nigrovittella, 106 Indian paintbrush, scarlet, 118 Indian skipper, 86 Indian wild rice, 123 Indiana myotis, 128 Indigo bunting, 56 Indigo blue wild, 117 cream wild, 117 yellow wild, 117 Inland silverside, 126 Insecta, 20, 105-107 Insects, 125, 130 air breathing, control of, 166-167 Iris fulva, 120 I. pseudacorus, 27 I. versicolor, 23, 26, 27 Iris blue water, 23, 26, 27 copper, 120 yellow water, 27 Ironoquia punctatissima, 21 Ironweed, New York, 123

JAMES, MICHAEL A., 133 Jay, blue, 54 Joan's swallowtail, 87 Joe-pye-weed spotted, 119 Steele's, 119 Johnny darter, 126 Jointed rush, 120

Isoetes butleri, 120

I. melanopoda, 120

Isopoda, 22

Isoperla holochlora, 20

Ixobrychus exilis, 127

Isopod, Clifton cave, 124

Ivory-billed woodpecker, 130

IONES, BRITTNEY, 1 JONES, SNAKE C., 165, 166 JUETT, BEVERLY W., 168 Juglans cinerea, 120 Junco hyemalis, 50 J. hyemalis, 53, 127 Junco, dark-eyed, 50, 53, 55, 127 Juncus articulatus, 120 I. elliottii, 120 I. filipendulus, 120 June, 120 Juniper, 87 ground, 120 Juniperus communis var. depressa, 120 Junonia coenia, 87 Juvenal's duskywing, 86

KALISZ, PAUL J., 1 Kentucky creekshell, 124 Kentucky lady's-slipper, 118 Kentucky red-backed vole, 128 KENTUCKY STATE NATURE PRESERVES COMMISSION, Kentucky vertebrate species, 171 Kidney-leaf grass-of-parnassus, 121 Kidney-leaf twayblade, 120 Kidneyshell, fluted, 124 King rail, 128 Kingfisher, belted, 69 Kinglet, golden-crowned, 55 Kingsnake, scarlet, 127 Kirchneriella lunaris, 34, 35 K. obesa, 34, 35 Kirtland's snake, 127 Mississippi, 127 swallow-tailed, 130 Koeleria macrantha, 120 KOGER, MATTHEW E., 169 Kricogonia lyside, 87 Krigia occidentalis, 120 KUMAR, K. S., 169

Lace-winged Roadside Skipper, 86 LACKI, MICHAEL J., 50 Ladies'-tresses Great Plains, 122 shining, 122 sweetscent, 122 yellow nodding, 122 Lady's-slipper Kentucky, 118 showy, 119 small white, 118 small yellow, 119 Lady American, 87 painted, 87 Laetilia fiskeella, 107 LAINE, SEPPO, 170

Lake chubsucker, 126

Lake cress, 117

Lake sturgeon, 125 Lampetra appendix, 126 Lamprey chestnut, 126 mountain brook, 126 northern brook, 126 southern brook, 126 Lampropeltis triangulum elapsoides, Lampsilis abrupta, 124 L. ovata, 124 Large bur-reed, 122 Large orange sulphur, 87 Large sedge, 118 Large spotted St. John's-wort, 120 Largeleaf grass-of-parnassus, 121 Lark sparrow, 127 Larkspur, Carolina, 119 Lasmigona compressa, 124 L. subviridis, 124 Lathyrus palustris, 120 L. venosus, 120 LAU, JOANN M., 168 Layside sulphur, 87 Lead, effects of on rats, 108-114 Leafcup, Tennessee, 121 Leafshell, 130 Cumberland, 130 Leafwing; goatweed, 87 Least bittern, 127 Least darter, 130 Least flycatcher, 127 Least madtom, 126 Least shrew, 164 Least skipper, 86 Least tern, 128 Least trillium, 123 Ozark, 123 Least weasel, 128 Leather-flower, blue jasmine, 118 Leavenworthia exigua var. laciniata, 120 L. torulosa, 120 Leiophyllum buxifolium, 120 Lemming, southern bog, 163, 164 Lenthus sp., 21 Leonard's skipper, 86 Leopard frog, northern, 127 Lepidoptera, 105–107 Lepidostoma sp., 21 Lepidostomatidae, 21 Lepomis macrochirus, 75 L. macrochirus, 100 L. marginatus, 126 L. miniatus, 126 Lepotes marina, 87 Leptochloa fascicularis, 167 Leptodea leptodon, 130 Leptophlebiid mayfly, 125

Leptophlebiidae, 20

Lerema accius, 86

Lerodea eufala, 86

Leptoxis praerosa, 123

Lescur's bladderpod, 120

ž		
Lespedeza capitata, 120	Gattinger's, 120	northern, 126
L. stuvei, 120	Nuttall's, 120	slender, 126
Lesquerella globosa, 120	Locust	Magnolia acuminata, 54
L. lescurii, 120	black, 54, 56	Maianthemum canadense, 120
Lesquereux's bladderpod, 120	water, 119	M. stellatum, 120
Lesser Adams cave beetle, 125	Loesel's twayblade, 120	Maindenhair fern, southern, 117
Lettuce-leaf saxifrage, 122	Logperch, 70	Malirekus hastatus, 20
Leucania calidior, 107	blotchside, 130 Lolium perenne, 167, 168	Mallow, hispid false, 120
Leucothoe recurva, 120 Leucrocuta, 15	Long-bract green orchis, 118	MALPHRUS, BENJAMIN K., 133 Malus angustifolia, 120
L. prob. thetis, 20	Long-eared owl, 127	Malvastrum hispidum, 120
Leuctra, 10, 13, 15	Long-styled rush, 120	Mammals, 128, 130
L. sp., 20	Long-tailed shrew, 128	Mammoth Cave shrimp, 124
Leuctridae, 20	Long-tailed skipper, 86	Mandarin, nodding, 119
Lexingtonia dolabelloides, 124	Longhead darter, 126	Manna grass
Liatris cylindracea, 120	Longhorn beetle, sixbanded, 125	pale, 123
Libythaena carinenta bachmanii, 87	Longleaf stitchwort, 122	sharp-scaled, 119
Lichens, 117	Longnose dace, 126	Manophylax butleri, 125
Lilium philadelphicum, 120	Longsolid, 124	Maple
L. superbum, 120	Lonicera dioica var. orientalis, 120	red, 54, 56
Lilliput	L. reticulata, 120	sugar, 54
purple, 124	Loosestrife	Mapleleaf, winged, 130
Texas, 124	Fraser's, 120	Marble, Olympia, 87
Lily	trailing, 120	Marigold, marsh, 130
calla, 27	Lophodytes cucullatus, 127 Lordithon niger, 125	Marine blue, 87
Turk's cap, 120 wood, 120	Lordithon rove beetle, black, 125	MARKER, GLENDA, 168 MARKEY, M., 170
Lily-of-the-valley	Lota lota, 126	Marsh marigold, 130
American, 118	Louisiana broomrape, 121	Marsh-pink, slender, 122
wild, 120	Louisville cave beetle, 125	Marshallia grandiflora, 120
Limenitis archippus, 87	Louisville crayfish, 124	Maryland glyph, 123
L. arthemis arthemis, 87	Lousewort, swamp, 121	Masked shrew, 128
L. arthemis astyanax, 87	Low rough aster, 117	Masticophis flagellum flagellum, 130
Limestone cave beetle, 125	Low-tech teaching methods, 170	Matelea carolinensis, 120
Limnephilid caddisfly, 125	open mindedness to, 170	Mathematics, 170
Limnephilidae, 21	Lucy Braun's white snakeroot, 117	Matricary grapefern, 117
Limnobium spongia, 120	Ludwigia hirtella, 120	Matted feather moss, 117
Limnophila sp., 22	Ludwigia, hairy, 120	Mayfly
Limnophyes sp., 22	Lumbriculidae, 1–5	burrowing, 125
Limstone fameflower, 122	Lumbriculidae, 20 Lumbricus, 3	ephemerellid, 125
Lip tern	Luperina trigona, 107	heptageniid, 125
Alabama, 118 Fèe's, 118	Lycaena hyllus, 87	leptophlebiid, 125 robust pentagenian burrowing,
Liparis loeselii, 120	Lycaena phlaeas americana, 87	130
Liriodendron tulipifera, 11, 54	Lycopodiella appressa, 120	McCALL, MAUREEN, 168
Listera australis, 120	L. inundata, 120	McDONOUGH, E., 170
L. smallii, 120	Lycopodium clavatum, 120	Meadow fritillary, 87
Lithasia armigera, 123	Lygropia tripunctata, 106	Meadow vole, 164
L. geniculata, 123	Lype diversa, 21	Meadow-parsnip, cutleaf, 123
L. salebrosa, 123	Lysimachia fraseri, 120	Meadowsweet, narrow-leaved, 122
L. verrucosa. 123	L. radicans, 120	Megaceryle alcyon, 71
Litobrancha recurvata, 125	L. terrestris, 120	Megalitter, 171
Little blue heron, 127	Lytrosis permagnaria, 125	an Appalachian deformity, 171
Little glassywing, 86		Megisto cymela, 87
Little spectaclecase, 124	Macrhybopsis gelida, 126	Melampyrum lineare var. latifolium,
Little wood satyr, 87	M. meeki, 126	M lineare war peotinatum 190
Little yellow, 87	Macrobrachium ohione, 124	M. lineare var. pectinatum, 120 Melanostoma, 155
Littlewing pearlymussel, 124 Lizard, eastern slender glass, 127	M. rosenbergii, 165–167	Melanthera nivea, 120
Lobelia appendiculata var. gattin-	Macroclemys temminckii, 127 Macroinvertebrate communities.	Melanthium parviflorum, 120
geri, 120	10–22	M. virginicum, 120
L. flabellaris, 27	Madtom	M. woodii, 120
L. nuttallii, 120	brown, 126	Meleagris gallopavo, 53
Lobelia	least, 126	Melithreptus, 155

Menidia beryllina, 126	first observations, 133–145	N-arginine dibasic convertase, 171
Mercury spurge, 119	MORGAN, ANN M., 166Morone	stability of acidic domain of, 171
Merganser, hooded, 127	chrysops × M. saxatilis, 166	Naiad, thread-like, 121
Meropleon cosmion, 107	Morus rubra, 54	Naididae, 20
Mesomphix rugeli, 123	MORZILLO, ANITA T., 164	Najas gracillima, 121
Metalmark	Mosses, 117	Nannyberry, 30–33
northern, 87	Moth	Narrow-leaved bluecurls, 123
swamp, 87, 125	geometrid, 125	Narrow-leaved meadowsweet, 122
Mice, radioprotective drug combi-	rattlesnake-master borer, 125	Nastra ilherminier, 86
nation in, 167–170	Moths, Kentucky, 105–107	Nathalis iole, 87
Michaux's bluets, 119	Mottled duskywing, 86	Neargyractis slossonalis, 106
Michaux's saxifrage, 122		Neckera pennata, 117
- Contract of the contract of	Mougeotia boodlei, 36, 38	
Micrasterias americana, 40, 43	M. sphaerocarpa, 36, 38	Necklace glade cress, 120
M. apiculata, 43, 44	Mountain brook lamprey, 126	Nemophila aphylla, 121
M. denticulata, 43, 44	Mountain creekshell, 124	Nemouridae, 20
M. laticeps, 43, 44	Mountain maple, 117	Neohelix dentifera, 123
M. pinnatifida, 43, 44	Mountain woodsia, 123	Neophylax, 15
M. truncata, 44	Mountain-lover, Canby's, 121	Neophylax sp., 21
Micropsectra sp., 22		Nephopterix crassifasciella, 107
	Mountain-mint	N. vesustella, 106
Microspora pachyderma, 36, 38	blunt, 121	
Microsporales, 36	white leaved, 121	Nephrocytium obesum, 34, 44
Microtendipes pedellus gp., 22	Mourning cloak, 87	Nerodia cyclopion, 127
M. rydalensis gp., 22	Mourning dove, 53	N. erythrogaster neglecta, 127
Microtus ochrogaster, 163, 164	Mouse	N. fasciata confluens, 127
M. pennsylvanicus, 164	cotton, 128	Nest density, artificial, 46–49
M. pinetorum, 162, 164		effect on Canada Goose, 46-49
Midland smooth softshell, 127	harvest, 163, 164	Nestronia umbellula, 121
	house, 164, 171	
Milbert's tortoise shell, 87	Moxostoma lacerum, 130	Net-spinning caddisfly, Helma's, 125
Milkwort	M. poecilurum, 126	Netrium digitus, 36, 39
cross-leaf, 121	M. valenciennesi, 130	Nettle-leaf noseburn, 123
Nuttall's, 121	Mucket, pink, 124	Nettle-leaf sage, 122
racemed, 121	Mud snake, western, 127	New available names, moths, 105–
Mimulus floribundus, 96		107
Minnow	Mud-plantain	New York ironweed, 123
cypress, 126	blue, 119	NEWCOMB, ANGELA L., 169
plains, 126	grassleaf, 119	Nicrophorus americanus, 125
	Muddy rocksnail, 123	
stargazing, 126	Mudminnow, central, 126	Night-heron
Minuartia cumberlandensis, 120	Muhlenbergia bushii, 120	black-crowned, 127
M. glabra, 120	M. cuspidata, 120	yellow-crowned, 127
Mirabilis albida, 120	M. glabriflora, 120	Nigronia, 14
Mississippi green water snake, 127	<u>.</u>	N. fasciatus, 21
Mississippi kite, 127	Muhly	Nilotanypus sp., 22
Missouri arrow-wood, 123	Bush's, 120	Nitrobacter, 27
Missouri rock-cress, 117	plains, 120	Nitrosomonas, 27
Mniotiilta vara, 53, 56	MURPHY, D. SHANNON, 133	Nocomis biguttatus, 126
	Mus musculus, 164, 171	
MOBLEY, JOEL, 169	Muscidae, 155	Noctuidae, 105, 107
Mock bishop's-weed, 121	Mussel	Nodding ladies'-tresses, yellow, 122
eastern, 121		Nodding mandarin, 119
Nuttall's, 121	oyster, 124	Nodding rattlesnake-root, 121
Mock orange, 121	salamander, 124	Northern bog club-moss, 120
hoary, 121	Mussels, freshwater, 124, 130	Northern broken-dash, 86
Molanna, 15, 18	Mustela nivalis, 128	Northern brook lamprey, 126
M. blenda, 21	Mycteria americana, 116	Northern cardinal, 53, 56
Molannidae, 21	Myotis austroriparius, 128	Northern cavefish, 125
Molothrus ater, 50, 54	M. grisescens, 128	Northern cloudywing, 86
Monarch, 87	M. leibii, 128	Northern coal skink, 127
Monarda punctata, 120	M. sodalis, 128	Northern crawfish frog, 127
Monkshood, blue, 117	Myotis	Northern dropseed, 122
Monotropsis odorata, 120	eastern small-footed, 128	Northern fox grape, 123
Montain birds, long-term conserva-	gray, 128	Northern hairstreak, 87, 125
tion, 50–59	Indiana, 128	Northern harrier, 127
Moorhen, common, 127	southeastern, 128	Northern leopard frog, 127
MOORMAN, KENNETH M., 169	Myriophyllum heterophyllum, 121	Northern madtom, 126
	, , , , , , , , , , , , , , , , , , , ,	
Morehead radio telescope, 133–145	M. pinnatum, 121	Northern metalmark, 87

O. solitaria, 34, 35

Northern pearly-eye, 87 Northern pike, 75 Northern pine snake, 127 Northern riffleshell, 124 Northern shoveler, 127 Northern starflower, 123 Northern starhead topminnow, 126 Northern white-cedar, 123 Northern witch, 119 Noseburn, nettle-leaf, 123 NOTES, 163-164 Notropis sp., 126 N. albizonatus, 126 N. hudsonius, 126 N. maculatus, 126 Noturus exilis, 126 N. hildebrandi, 126 N. phaeus, 126 N. stigmosus, 126 Nut-rush, fringed, 122 Nuthatch, red-breasted, 55, 128 Nuttall's lobelia, 120 Nuttall's milkwort, 121 Nuttall's mock bishop's-weed, 121 Nyctanassa violacea, 127 Nycticeius humeralis, 128 Nycticorax nycticorax, 127 Nyctiphylax sp., 21 Nymphalis antiopa, 87 Nymphalis vaualbum j-album, 87 Nyssa sylvatica, 54

Obovaria retusa, 124 Ocola skipper, 86 Odonata, 21 Oecophoridae, 105 Oedogoniales, 36 Oedogonium boscii, 36, 38 O. capilliforme, 36, 38 O. cardiacum, 36, 38 O. grande, 36, 38 Oenothera linifolia, 121 O. oakesiana, 121 O. perennis, 121 O. triloba, 121 Ohio shrimp, 124 Oidaematophorus eupatorii, 107 Old Well Cave beetle, 125 Oldenlandia uniflora, 121 Olethreutes tiliana, 106 Oligia mactata, 107 Oligochaeta, 1-5, 20 Olivaceous sedge, 119 Olive darter, 126 Olympia marble, 87 Oncophorus raui, 117 One-flower fiddleleaf, 119 Onosmodium molle ssp. hispidissimum, 121 O. molle ssp. molle, 121 O. molle ssp. occidentale, 121 Onychonema laeve, 44

Onyx rocksnail, 123

Oocystis parva, 34, 35

Ophiogomphus aspersus, 125 O. howei, 125 Ophisaurus attenuatus longicaudus, Ophorornis formosus, 53 Orange sulphur, 87 Orange tip, falcate, 87 Orange, hoary mock, 121 Orange, mock, 121 Orange, sleepy, 87 Orange-barred sulphur, 87 Orangefood pimpleback, 124 Orbesilum stipulatum, 130 Orchard oriole, 53 Orchid small purple-finged, 121 white fringeless, 121 yellow-crested, 121 Orchis, long-bract green, 118 Orconectes burri, 124 O. inermis, 124 O. jeffersoni, 124 O. lancifer, 124 O. palmeri, 124 O. pellucidus, 124 Oriole, orchard, 53 Ornate rocksnail, 123 Orobanche ludoviciana, 121 Oronectes australis, 124 O. bisectus, 124 Orontium aquaticum, 121 Orthocladius annectens, 22 Orthocyclops modestus, 165-166 Orthotrichum diaphanum, 117 Osprey, 128 Ostrocerca prob. truncata, 20 Oulimnious latiusculus, 21 Ovate catchfly, 122 Ovate fiddleleaf, 119 Ovenbird, 53 Owl barn, 128 feeding habits, 163-164 long-eared, 127 short-eared, 127 Oxalis priceae, 121 Oxydendrum arboreum, 54 Oyster mussel, 124 Ozark least trillium, 123 Paintbrush, scarlet Indian, 118 Painted lady, 87 Painted trillium, 123 Painted turtle, southern, 127 Palaemonias ganteri, 124 Palamedes swallowtail, 87

PALAZZOLO, D. L., 169 Pale false foxglove, 117 Pale manna grass, 123 Pale umbrella-wort, 120 Palezone shiner, 126 Pallid shiner, 126

PALOMBI, PEGGY SHADDUCK, 169 Palpomyia sp., 21 Pale corydalis, 118 Pandion haliaetus, 128 PANEMANGALORE, MYNA, 108 Panoquina ocola, 86 Papaipema eryngii, 125 Papershell, Cumberland, 124 Papilio cresphontes, 87 P. joanae, 87 P. palamedes, 87 P. polyxenes asterius, 87 P. troilus, 87 Papilo glaucus, 87 Parachaetocladius sp., 22 Paragnetina sp., 20 Parakeet, Carolina, 130 Paraleptophlebia, 10, 13, 15 P. prob. ontario, 20 Parametriocnemus, 15 P. lundbecki, 22 Parapediasia decorella, 106 Parnassia asarifolia, 121 P. grandifolia, 121 Paronychia argyrocoma, 121 Parrhasius m-album, 87 Parsley, prairie, 130 Parus bicolor, 53 Parus carolinensis, 53 Paspalum, bull, 121 Paspalum boscianum, 121 Passenger pigeon, 130 Passerculus sandwichensis, 128 Passerina cyanea, 53, 56 Pastinaca sativa, 159 Patera panselenus, 123 Pawpaw, 165, 166 controlled crosses, 165 Kentucky State University project, 166 molecular markers, 165 Paxistima canbyi, 121 Peachleaf willow, 122 Peacock, white, 87 Pearl crescent, 87 Pearly-eye Creole, 87 northern, 87 southern, 87 Pearlymussel cracking, 130 dromedary, 130 littlewing, 124 slabside, 124 Peatmoss, 117 smooth veiny, 120 vetchling, 120 Peck's skipper, 86 Pediastrum boryanum, 36, 37

P. duplex, 36, 37

P. simplex, 36, 37

Pallid sturgeon, 126

Pedicularis lanceolata, 121	P. villosus, 53	Poa saltuensis, 121
Pegias fabula, 124	Pied-billed grebe, 128	Poaceae, 88
Peltoperla arcuata, 20	Pieris rapae, 87	Poanes hobomok, 86
Peltoperlidae, 20	P. virginiensis, 87	P. viator, 86
Penium margaritaceum, 36, 39	Pigeon, passenger, 130	P. yehl, 86
Pennant, double-ringed, 125	Pigmy rattlesnake, western, 127	P. zabulon, 86
Pennyroyal, rough, 119	Pigtoe	Pocketbook, 124
Pentagenia robusta, 130	pyramid, 124	fat, 124
Pentagenian burrowing mayfly, ro-	rough, 124	Podilymbus podiceps, 128
bust, 130	Pike, northern, 75	Podostemum ceratophyllum, 121
Pepper and salt skipper, 86	Pilaria sp., 22	Pogonia, rose, 121
Percina burtoni, 130	Pilsbryna sp., 123	Pogonia ophioglossoides, 121
P. caprodes, 70	Pimpleback, orangefoot, 124	Poison sumac, 123
P. copelandi, 70	Pine Mountain tigersnail, 123	Polioptila caerulea, 53
P. macrocephala, 126	Pine snake, northern, 127	Polites origenes, 86
P. squamata, 126	Pine, Virginia, 55	P. peckius, 86
Percopsis omiscomaycus, 126	Pines, 52	P. themistocles, 86
Peregrine falcon, 127	Pinesap, sweet, 120	Polycentropodidae, 21
Perideridia americana, 121	Pink mucket, 124	Polycentropus sp., 21
Perlidae, 20	Pink, ring, 124	Polydamas swallowtail, 87
Perlodidae, 20	Pinus virginiana, 52, 54, 55	Polygala cruciata, 121
Peromyscus gossypinus, 128	Pipevine swallowtail, 86	P. nuttallii, 121
Pesticide residue, 165	Pipilo erythrophthalmus, 53, 54	P. paucifolia, 121
in soil, 165	Piranga olivacea, 53	P. polygama, 121
measurement, 165	Pisidium sp., 20	Polygonia comma, 87
mitigation, 165	Pituophis melanoleucus melanoleu-	P. faunus, 125
runoff, 165	cus, 127	P. faunus smythi, 87
Phacelia ranunculacea, 121	Plains frostweed, 119	P. intearrogationis, 87
Phaeophyscia leana, 117	Plains minnow, 126	P. progne, 87, 125
Phalacrocorax auritus, 128	Plains muhly, 120	Polymnia laevigata, 121
Phalaris arundinacea, 23, 26, 27	Planariidae, 20	Polypedilum aviceps gp., 22
Phenacobius uranops, 126	Planktosphaeria gelatinosa, 34, 35	P. convictum gp., 22
Pheucticus ludovicianus, 50	Plant oils, effectiveness on air	P. fallax gp., 22
P. ludovicianus, 53, 128		
	breathing insects, 166–171	P. haltarare gp., 22
Philadelphus inodorus, 121	Plantago cordata, 121	Polytaenia nuttalli, 130
P. pubescens, 121	P. lanceolata, 167, 168	Polytrichum pallidisetum, 117
PHILLIPS, B., 170	Plantain, heartleat, 121	P. piliferum, 117
Philopotamidae, 21	Plants, 117, 130	P. strictum, 117
Philtraea monillata, 107	Platanthera cristata, 121	Pompeius verna, 86
Phlox bifida ssp. bifida, 121	P. integrilabia, 121	POMPER, KIRK W., 165, 166
P. bifida ssp. stellaria, 121	P. psycodes, 121	POND, GREGORY J., 10
Phlox	Platygobio gracilis, 126	Pondweed
cleft, 121	Plecoptera, 20	Illinois, 121
starry cleft, 121	Plethadon hoffmani, 8	spotted, 121
Phoebe, eastern, 53	P. wehrlei, 8	Pontederia cordata, 23, 26, 27, 121
Phoebis agarithe, 87	Plethobascus cicatricosus, 130	Pontia protodice, 87
P. philea, 87	P. cooperianus, 124	Pooecetes gramineus, 128
P. sennae, 87	P. cyphyus, 124	Poppy-mallow, clustered, 118
Pholisora catullus, 86	Plethodon cinereus, 6–9, 127	Porcupine sedge, 118
Photoacoustic measurements, 169	P. electromorphus, 6	Porter's reed 118
in biological fluids, 169	P. richmondi, 6–9	Possum haw viburnum, 123
in biological tissues, 169	P. shenandoah, 8	Potamilus capax, 124
Phoxinus cumberlandensis, 126	P. wehrlei, 127	Potamilus purpuratus, 124
Phyciodes batesii, 87, 125	Pleurobema clava, 124	Potamogeton illinoensis, 121
P. tharos, 87	P. oviforme, 124	P. pulcher, 121
Physics & Astronomy, 170	P. plenum, 124	Potato-bean, Price's, 117
Physiology & Biochemistry, 170–172	P. rubrum, 124	Potentilla erecta, 158
Physostegia intermedia, 130	Pleurocera alveare, 123	Potthastia sp., 22
Picea rubens, 52	P. curta, 123	Poultry by-product meal, 166
Pickerel weed, 23, 26, 27	Pleurocercidae, 60–63	in diets for fish, 166
Pickerel, chain, 126	Pleurotaenium constrictum, 39, 40	Prairie gentian, 119
Pickerel-weed, 121	P. ehrenbergii, 39, 40	Prairie parsley, 130
Picoides borealis, 128	P. nodosum, 39, 40	Prairie redroot, 118
P. pubescens, 53	Plukenet's cyperus, 118	Prairie vole, 161, 164

Prairie-chicken, greater, 130 Prairie-clover, purple, 119 Prawns, freshwater, 165, 166 copepods as live food for, 165-166 effect of water temperature on survival, 167 transport density on survival, 165 Pre-emphasis filter, auditory system model, 169 Prenanthes alba, 121 P. aspera, 121 P. barbata, 121 P. crepidinea, 121 Pretty St. John's-wort, 120 Price's potato-bean, 117 Price's yellow wood sorrel, 121 Prickly bog sedge, 118 Primrose evening, 121 yellow evening, 118 Pristina aequisita, 20 Privet, upland, 119 Procambarus viaeviridis, 125 Prosimulium sp., 22 Prosopis, 155 Prostate cancer cell lines, 170-171 Proterometra albacauda, 103 P. autraini, 102 P. catenaria, 103 P. dickermani, 102 P. edneyi, 60-63, 103 P. macrostoma, 99-104 development in host, 99-104 distome emergence, 99-104 P. sagittaria, 103 P. septimae, 103 Prunus serotina, 54 Psephenidae, 21 Psephenus herricki, 21 Pseudanophthalmus audax, 125 P. calcareus, 125 P. catoryctos, 125 P. conditus, 125 P. desertus major, 125 P. frigidus, 125 P. globiceps, 125 P. horni abditus, 125 P. horni caecus, 125 P. horni horni, 125 P. hypolithos, 125 P. inexpectatus, 125 P. parvus, 125 P. pholeter, 125 P. pubescens intrepidus, 125 P. puteanus, 125 P. rogersae, 125 P. scholasticus, 125 P. simulans, 125 P. tenebrosus, 125 P. troglodytes, 125 Pseudolimnophila sp., 22 Pseudoroegneria spicata, 92 Pseudostenophylax uniformis, 21

Psoralidium tenuiflorum, 121 Pterophoridae, 105, 107 Ptilimnium capillaceum, 121 P. costatum, 121 P. nuttallii, 121 Ptychobranchus subtentum, 124 Puma concolor couguar, 130 Puma, eastern, 130 Punctate coil, 123 Purple giant hyssop, 117 Purple lilliput, 124 Purple prairie-clover, 119 Purple sand grass, 123 Purple, red spotted, 87 Purple-fringed orchid, small, 121 Purple-oat, 122 Pussy willow, 122 Pycnanthemum albescens, 121 P. muticum, 121 Pycnopsyche gentilis, 21 Pycnopsyche prob. guttifer, 21 Pygmy snaketail, 125 Pyramid pigtoe, 124 Pyrausta signatalis, 106 Pyrgus centaurae, 86 P. communis, 86 P. wyandot, 125 Pyrola americana, 121

Quadrigula chodatii, 36, 37 Quadrula cylindrica cylindrica, 124 Q. fragosa, 130 Q. tuberosa, 130 Queen, 87 Queen crater, 123 Quercus alba, 11, 54 Q. coccinea, 11 Q. falcata, 54 Q. macrocarpa, 89 Q. mandanensis, 89 Q. montana, 11 Q. muehlenbergii, 54, 89 Q. prinojdes, 89 Q. prinus, 54 Q. rubra, 54 Question mark, 87 Quillwort blackfoot, 120 Butler's, 120

Rabbit-tobacco, small, 119
Rabbitsfoot, 124
Rabdotus dealbatus, 123
Rabdotus, whitewashed, 123
Racemed milkwort, 121
Radio telescope, Morehead, 133—145
Radioprotective drug combination in mice, 169—170
Rafinesque's big-eared bat, 128
Rail, king, 128
Rainbow darter, 70
Rallus elegans, 128
Rana areolata circulosa, 127

Rand's goldenrod, 122 Ranunculaceae, 64 Ranunculus ambigens, 121 Raptoheptagenia cruentata, 125 Rare biota of Kentucky, 115-132 adult, 108-114 aged, 108-114 body and tissue weights, 108-114 effects of cadmium on, 108-114 effects of lead on, 108-114 effects of zinc on, 108-114 weanling, 108-114 Rattlesnake, western pigmy, 127 Rattlesnake-master borer moth, 125 Rattlesnake-root barbed, 121 nodding, 121 rough, 121 white, 121 Raven, common, 127 Rayed bean, 124 Red admiral, 87 Red buckeye, 117 Red elderberry, 122 Red maple, 54, 56 Red spotted purple, 87 Red spruce, 52 Red turtlehead, 118 Red wolf, 130 Red-backed vole, Kentucky, 128 Red-banded, 87 Red-breasted nuthatch, 55, 128 Red-cockaded woodpecker, 128 Red-eyed vireo, 54, 55 Red-winged blackbird, 164 Redback salamander, 127 Redhorse blacktail, 126 greater, 130 Redroot, prairie, 118 Redspotted sunfish, 126 Redstart, American, 56 Reed bent grass, 118 Reed canary grass, 23, 26 Reed grass, blue-joint, 118 Porter's, 118 REED, EDDIE B., 166 REEDER, BRIAN C., 46 Regal fritillary, 87, 125 Regulus satrapa, 55 Reithrodontomys megalotis, 164 Reithrodontomys spp., 163 Relict darter, 126 Reniform sedge, 118 Reptiles, 127, 130 Rheotanytarsus sp., 22 Rhinichthys cataractae, 126 Rhodacme elatior, 123 Rhododendron canescens, 121 Rhyacionia aktita, 106 Rhyacophila carolina, 21 R. invaria gp., 21

Rana pipiens, 127

	index to volume of	10
R. torva, 21	Round-headed cave beetle, 125	S. liparops, 87
R. vibox, 21	Roundleaf fameflower, 123	S. titus mopsus, 87
Rhyacophilidae, 21	Rove beetle, black lordithon, 125	Satyrodes appalachia, 87
Rhynchosia tomentosa, 121	Royal catchfly, 122	Savannah sparrow, 128
Rhynchospora globularis, 121	Rubus canadensis, 122	Sawfin shiner, 126
R. macrostachya, 121	R. whartoniae, 122	Saxifraga michauxii, 122
Rhyngia, 155	Ruby-throated hummingbird, 53	S. micranthidifolia, 122
Ribbon snake	Rudbeckia subtomentosa, 122	S. pensylvanica, 122
eastern, 127	Rufous-sided towhee, 54, 56	Saxifrage
western, 127	Rugged hornsnail; 123	brook, 118
RICE, DAVID, 169	Running buffalo clover, 123	lettuce-leaf, 122
Rice	Running-pine, 120	Michaux's, 122
Indian wild, 123	Rush	swamp, 122
southern wild, 123	bog, 120	Sayornis phoebe, 53
Riffleshell	jointed, 120	Scaleshell, 130
angled, 130	long-styled, 120	Scallopwing, Hayhurst's, 86
northern, 124	Ruta graveolens, 159	Scaly sand darter, 130
tan, 130	Rye, Svenson's wild, 119	Scaphirhynchus albus, 126
Tennessee, 130		Scarlet indian paintbrush, 118
Wabash, 130	Sabatia campanulata, 122	Scarlet kingsnake, 127
Rigid sedge, 118	Sachem, 86	Scenedesmus bicaudatus, 36, 37
Ring pink, 124	Sage, nettle-leaf, 122	S. dimorphus, 36, 37
Riparia riparia, 128	Sagittaria graminea, 122	S. obtusus, 36, 37
River bulrush, 122	S. latifolia, 27	S. quadricauda, 36, 37
Roan Mountain goldenrod, 122	S. platyphylla, 122	Schisandra glabra, 122
Roan sedge, 118	S. rigida, 122	Schizachne purpurascens, 122
Robinia pseudoacacia, 54	Salamander mussel, 124	Scholarly cave beetle, 125
ROBINSON, DAVID L., 167, 168	redback, 127	Schroederia setigera, 34, 35
Robust pentagenian burrowing may-	three-lined, 126	SCHUSTER, JESSICA, 62, 99
fly, 130	Wehrle's, 127	Schwalbea americana, 122
Rock skullcap, 122	Salebriaria atratella, 106	Science Education, 171
Rock-cress	S. tenebrosella, 106	Scientists of Kentucky, 77–85
Braun's, 117	Salix amygdaloides, 122	Scirpus expansus, 122
hairy, 117	S. discolor, 122	S. fluviatilis, 122
Missouri, 117	S. nigra, 54	S. hallii, 122
Rockcastle aster, 117	Salvia urticifolia, 122	S. heterochaetus, 122
Rockshell, rough, 130	Sambucus racemosa ssp. pubens,	S. microcarpus, 122
Rocksnail	122	S. validus, 23, 26, 27
armored, 123	Sand darter	S. verecundus, 122
muddy, 123 onyx, 123	scaly, 130	Scleria ciliata var. ciliata, 122 Scorpion-weed, blue, 121
ornate, 123	western, 125	
varicose, 123	Sand grape, 123	Screwstem, yellow, 117
	Sand grass, purple, 123	Scrophulariaceae, 96
Roger's cave beetle, 125 Rosaceae, 146–162	Sand-myrtle, 120	Sculpted glyph, 123 Scurf-pea stipuled, 130
Rose pogonia, 121	Sandpiper	few-flowered, 121
Rose turtlehead, 118	spotted, 127 upland, 127	Scutellaria arguta, 122
Rose verbena, 119	Sandwort	S. saxatilis, 122
Rose-breasted grosbeak, 50, 53, 55,	Appalachian, 120	Sedge wren, 127
56, 128	Cumberland, 120	Sedge
Rosemary, Cumberland, 118	Sanguisorba canadensis, 122	Appalachian, 118
ROSEN, RONALD, 62, 99	Santhidium armatum, 40, 42	bristly, 118
Rosinweed, Appalachian, 122	Sapsucker, yellow-bellied, 53	broadwing, 118
Rosinweed, tansy, 122	Sassafras, 56	brown bog, 118
Rosy twistedstalk, 122	Sassafras albidum, 54, 56	cedar, 118
ROTH, LAUREN, 62	Satyr	Coastal Plain, 118
Rough aster, low, 117	Ćarolina, 87	Crawe's, 118
Rough dropseed, 122	gemmed, 87	cypress-swamp, 118
Rough pennyroyal, 119	little wood, 87	epiphytic, 118
Rough pigtoe, 124	Satyrium acadicum, 87	finely-nerved, 118
Rough rattlesnake-root, 121	S. calanus falacer, 87	Fraser's, 118
Rough rockshell, 130	S. caryaevorum, 87	large, 118
Round combshell, 130	S. edwardsii, 87	olivaceous, 119
Round-head bush-clover, 120	S. favonius ontario, 87, 125	porcupine, 118

prickly bog, 118	S. pinnatifidum, 122	Sleepy orange, 87
reniform, 118	S. wasiotense, 122	Slender blazingstar, 120
rigid, 118	Silver-bordered fritillary, 87	Slender bulrush, 122
roan, 118	Silver-spotted skipper, 86	Slender dragon-head, 130
stalkgrain, 118	Silverbell, common, 119	Slender glass lizard, eastern, 127
straw, 118	Silvering, 121	Slender madtom, 126
summer, 118	Silvery aster, silvery, 117	Slender marsh-pink, 122
Tarheel, 118	Silvery blue, 87	Small enchanter's-nightshade, 118
umbel-like, 118	Silvery checkerspot, 87	Small lady's-slipper, yellow, 119
weak stellate, 118	Simpsonaias ambigua, 124	Small purple-fringed orchid, 121
woolly, 118	Simuliidae, 22	Small rabbit-tobacco, 119
Sedum telephioides, 122	Simulium sp., 22	Small sundrops, 121
SEED, T. M, 169	Sistrurus miliarius streckeri, 127	Small white lady's-slipper, 118
Seiurus aurocapillus, 53	Sitanion hystrix, 88, 89, 92	Small yellow lady's-slipper, 119
Selenastrum gracile, 34, 35	S. minus, 88	Small-flower baby-blue-eyes, 121
	and the second s	
Selenisa sueroides, 107	Sitta canadensis, 55, 128	Small-flowered false hellebore, 120
Semotilus atromaculatus, 11	S. carolinensis, 53	Small-flowered thoroughwort, 119
September elm, 123	Sixbanded longhorn beetle, 125	Small-footed myotis, eastern, 128
Sessile-fruit arrowhead, 122	Skeleton grass	Small-fruit bulrush, 122
Setophaga ruticilla, 53, 56	bearded, 119	Smallscale darter, 126
Shad, Alabama, 125	shortleaf, 119	SMITH, J., 171
Shaggy cavesnail, 123	Skink	Smooth blackberry, 122
Shaggy hedge-hyssop, 119	northern coal, 127	Smooth softshell, midland, 127
Sharp-scaled manna grass, 119	southeastern five-lined, 127	Smooth veiny peavine, 120
Sharp-shinned hawk, 127	southern coal, 127	Snail, 60–63, 121
Shawnee darter, 126	Skipper	Snake
Sheepnose, 124	Appalachian grizzled, 125	broad-banded water, 127
Shell galaxies, hydrogen gas in, 170	Bell's roadside, 86	copperbelly water, 127
SHIBER, JOHN G., 170	broadwinged, 86	corn, 127
Shiner	clouded, 86	eastern ribbon, 127
blacktail, 125	cobweb, 86	Kirtland's, 127
bluntface, 125	common checkered, 86	Mississippi green water, 127
palezone, 126	common roadside, 86	northern pine, 127
pallid, 126	crossline, 86	western mud, 127
sawfin, 126	Delaware, 86	western ribbon, 127
spottail, 126	Dion, 86	Snakeroot
taillight, 126	Duke's, 86, 125	Lucy Braun's white, 117
Shining ladies'-tresses, 122	Dun, 86	white, 167
Shooting-star, French's, 119	dusted, 86	Snaketail
Short's goldenrod, 122	Eufala, 86	brook, 125
Short's hedge-hyssop, 119	European, 86	pygmy, 125
Short-eared owl, 127	fiery, 86	Snapping turtle, alligator, 127
Shortleaf skeleton 119	gold-banded, 86	Snout-bean, hairy, 121
Shortspire hornsnail, 123	grizzled, 86	Snow melanthera, 120
and the second second	hobomok, 86	Snow trillium, 123
Shoveler, northern, 127 Showy gentian, 119	Indian, 86	Snowberry, 122
Showy lady's-slipper, 119		Snuffbox, 124
Shrew, 163	lace-winged roadside, 86	Soft false gromwell, 121
·	least, 86	Soft-haired thermopsis, 123
least, 164	Leonard's, 86	
long-tailed, 128	long-tailed, 86	Softshell, midland smooth, 127
masked, 128	Ocala, 86	Softstem bulrush, 23, 26, 27
Shrimp	Peck's, 86	Solidago albopilosa, 122
Mammoth Cave, 124	pepper and salt, 86	S. buckleyi, 122
Ohio, 124	silver-spotted, 86	S. curtisii, 122
Sicklefin chub, 126	swarthy, 86	S. gracillima, 122
Sida hermaphrodita, 122	tawny-edged, 86	S. puberula, 122
Side-oats grama, 118	Yehl, 86	S. roanensis, 122
Silene ovata, 122	Zabulon, 86	S. shortii, 122
S. regia, 122	Skullcap	S. simplex ssp. randii, 122
Silky aster, barrens, 117	hairy, 122	S. squarrosa, 122
Silphium sunflower, 119	rock, 122	Solomon-seal, starry false, 120
Silphium laciniatum var. laciniatum,	Skunk, eastern spotted, 128	Sootywing, common, 86
122	Slabside pearlymussel, 124	Sorastrum americanum, 36, 37
S. laciniatum var. robinsonii, 122	Sleepy duskywing, 86	Sorex spp., 163
,		* *

S. cinereus, 128	Spottail shiner, 126	Straw sedge, 118
S. dispar blitchi, 128	Spotted beebalm, 120	Streptopus roseus var. perspectus,
Sorrel, Price's yellow wood, 121	Spotted coralroot, 118	122
Southeastern five-lined skink, 127	Spotted darter, 126	Striped bass hybrid, 166
Southeastern myotis, 128	Spotted joe-pye-weed, 119	Striped hairstreak, 87
Southern bog club-moss, 120	Spotted pondweed, 121	Striped whitelip, 124
Southern bog goldenrod, 122	Spotted sandpiper, 127	Strymon melinus, 87
Southern bog lemming, 163	Spotted skunk, eastern, 128	Sturgeon chub, 126
Southern broken-dash, 86	Spreading false foxglove, 117	Sturgeon
Southern brook lamprey, 126	Spring azure, 87	lake, 125
Southern cavefish, 126	Spruce, red, 52	pallid, 126
Southern cloudywing, 86	Spurge, mercury, 119	Stygobromus vitreus, 125
Southern coal skink, 127	Squarrose goldenrod, 122	Stylogomphus, 14
Southern crabapple, 120	SŘINIVASÁN, V., 169	S. albistylus, 21
Southern dogface, 87	St. John's-wort	Stylurus notatus, 125
Southern heartleaf, 119	creeping, 119	Sucker
Southern maidenhair fern, 117	large spotted, 120	blackfin, 126
Southern painted turtle, 127	pretty, 120	hairlip, 130
Southern pearly-eye, 87	St. Peter's-wort, 119	Sugar maple, 54
Southern shield wood fern, 119	Stachys eplingii, 122	Sugarspoon, 130
Southern twayblade, 120	Stalkgrain sedge, 118	Sulphur
Southern wild rice, 123	Staphylus hayhurstii, 86	clouded, 87
Soyedina sp., 20	Star tickseed, 118	cloudless, 87
Sparganium eurycarpum, 120	Starflower, northern, 123	dainty, 87
Sparrow	Stargazing minnow, 126	large orange, 87
Bachman's, 127	Starhead topminnow, northern, 126	Layside, 87
Henslow's, 127	Starry cleft phlox, 121	orange, 87
lark, 127	Starry false solomon-seal, 120	orange-barred, 87
Savannah, 128	Starvine, bay, 122	Sumac, poison, 123
vesper, 128	State records, moths, 105–107	Summer sedge, 118
Spearwort, water-plantain, 121	Staurastrum alternans, 40, 42	Sundew Sundew
Speckled darter, 70	S. arctiscon, 40, 42	dwarf, 119
Spectaclecase, 124	S. botryophilum, 40, 42	spoon-leaved, 119
little, 124	S. brasiliense, 40, 42	Sundrops
Speedwell, American, 123	S. chaetoceros, 40, 42	small, 121
Speyeria aphrodite, 87	S. crenulatum, 40, 42	thread-leaf, 121
S. cybele, 87	S. curvatum, 40, 42	Sunfish
S. diana, 87	S. dickiei, 40, 42	dollar, 126
S. idalia, 87, 125	S. hexacerum, 40, 42	redspotted, 126
Sphaeriidae, 20		Sunflower
	S. leptocladum, 40, 42 S. limneticum, 40, 42	
Sphaerocystis schroeteri, 34, 35		Eggert's, 119
Sphagroup guinguafarium, 44	S. setigerum, 40, 42	silphium, 119
Sphagnum quinquefarium, 117	Steele's joe-pye-weed, 119	Sunshine bass, 166 diets for, 166
Sphenopholis pensylvanica, 122	Stellaria fontinalis, 122	
Sphyrapicus varius, 53	S. longifolia, 122 Stollate sodge week, 118	Superficial coset curiosities, 170
Spicebush swallowtail, 87	Stellate sedge, weak, 118	Supplejack, 117
Spiked hoary-pea, 123	Stemless evening-primrose, 121	Surprising cave beetle, 125
Spilogale putorius, 128	Stenacron interpunctatum, 20	Svenson's wild rye, 119
Spinulose wood fern, 119	Stenelmis sp., 21	Swallow
Spiraea alba var. alba, 122	S. crenata, 21	bank, 128
S. virginiana, 122	Stenonema bednariki, 125	polydamas, 87
Spiraea, Virginia, 122	S. meririvulanum, 20	Swallow-tailed kite, 130
Spiranthes lucida, 122	S. meririvulanum, 15	Swallowtail
S. magnicamporum, 122	S. vicarium, 20	black, 87
S. ochroleuca, 122	Stenoptilodes brevipennis, 107	giant, 87
S. odorata, 122	Sterna antillarum, 128	Joan's, 87
Spirogyra communis, 36, 38	Stevens Creek Cave beetle, 125	palamedes, 87
S. pratensis, 36, 38	Stipuled scurf-pea, 130	pipevine, 86
S. varians, 36, 38	Stitchwort	spicebush, 87
Spirotaenia condensata, 36, 39	longleaf, 122	tiger, 87
Spondylosium moniliforme, 44	water, 122	zebra, 87
Spoon-leaved sundew, 119	Stonecrop, Allegheny, 122	Swamp darter, 126
Sporobolus clandestinus, 122	Stonefly, 18	Swamp lousewort, 121
S. heterolepis, 122	Stoneroller, central, 75	Swamp metalmark, 87

Swamp metalmark, 125 Swamp saxifrage, 122 Swamp wedgescale, 122 Swamp-candles, 120 Swarthy skipper, 86 Sweet birch, 54 Sweet coneflower, 122 Sweet flag, 23, 26, 27 Sweet pinesap, 120 Sweet-fern, 118 Sweetscent ladies'-tresses, 122 Sweetshrub, 118 Sweltsa, 15 S. sp., 20 Symphoricarpos albus, 122 Synaptomys cooperi, 163, 164 Synclita tinealis, 106 Synorthocladius sp., 22 Syritta, 155 Syrphidae, 155 Syrphus, 155 Taillight shiner, 126 Talinum calcaricum, 122 Tall beaked-rush, 121 Tall bush-clover, 120 Tall fescue, 27 Tall hairy groovebur, 117 Talnium teretifolium, 123 Tamoxifen, 170-171 171 Tan riffleshell, 130

effect on Cathepsin B, 170-171 effect on Cathepsin L, 170–171 effect on cysteine proteases, 170-Tansy rosinweed, 122 Tanytarsus, 15 Tanytarsus sp., 22 Taraxacum officinale, 167, 168 inheritance of morphological characteristics, 167 inheritance of physiological characteristics, 167 Tarheel sedge, 118 Tatum Cave beetle, 125 Tawny cotton-grass, 119 Tawny crescent, 87, 125 Tawny emperor, 87 Tawny-edged skipper, 86 Taxus canadensis, 123 Teal, blue-winged, 127 Teilingia excavata, 44 Telescope, Morehead radio, 133-Ten-lobe false foxglove, 117 Tennessee aster, 117 Tennessee clubshell, 124 Tennessee leafcup, 121 Tennessee riffleshell, 130 Tephrosia spicata, 123 Tern black, 130

Terrestrial vertebrate species, 171

least, 128

richness of, 171 Tetgrasporales, 34 Tetmemorus brebissonii, 39, 40 Tetraedron minimum, 34, 35 T. regulare, 34, 35 Texas aster, 117 Texas lilliput, 124 Thalictrum clavatum, 62 T. mirabile, 62 chromosome number of, 62-63 Thamnophis proximus proximus, T. sauritus sauritus, 127 Thaspium pinnatifidum, 123 Theliopsyche, 15, 18 T. sp., 21 Thermopsis mollis, 123 soft-haired, 123 Thienimanniella sp., 22 THIERET, JOHN W., 146 Thoburnia atripinnis, 126 THOGMARTÎN, WAYNE E., 164 THOMPSON, J., 170 THOMPSON, KENNETH R., 166 Thoroughwort, small-flowered, 119 Thorybes bathyllus, 86 T. confusis, 86 T. pylades, 86 Thread-leaf sundrops, 121 Thread-like naiad, 121 Threadfoot, 121 Three-awn grass, branched, 117 Three-lined salamander, 126 Three-toed amphiuma, 126 Thrush, wood, 56 Thryomanes bewickii, 128 Thryothorus ludovicianus, 53, 56 Thuja occidentalis, 123 Thymelicus lineola, 86 Tickseed, star, 118 TIDWELL, JAMES H., 165, 166, Tiger swallowtail, 87 Tigersnail, Pine Mountain, 123 Tilia spp., 54 Tillium nivale, 123 Tippecanoe darter, 70 Tipula sp., 22 Tipulidae, 22 Topminnow golden, 126 northern starhead, 126 Torreyochloa pallida, 123 Tortoise shell, compton, 87 Milbert's, 87 Tortricidae, 105, 106 Tortula, 117 Tortula norvegica, 117 Towhee, rufous-sided, 54, 56 Toxicodendron vernix, 123 Toxolasma lividus, 124

T. texasiensis, 124

Tragia urticifolia, 123

Trailing loosestrife, 120

Traverella lewisi, 125 Treatment, wastewater, 23-29 Treefrog barking, 127 bird-voiced, 126 gray, 127 green, 127 Trematode, digenetic, 60-63 Trepocarpus, 123 Trepocarpus aethusae, 123 Trichoptera, 21 Trichostema setaceum, 123 Tricladia, 20 Trientalis borealis, 123 Trifolium reflexum, 123 T. repens, 167, 168 T. stoloniferum, 92, 123 Trillium pusillum var. ozarkanum, T. pusillum var. pusillum, 123 T. undulatum, 123 Trillium least, 123 Ozark least, 123 painted, 123 snow, 123 Triplasis purpurea, 123 Troglodytes troglodytes, 55 Trout, American brook, 126 Trout-perch, 126 Tryphlichthys subterraneus, 126 Tsuga canadensis, 11 TUAN, VO-DINH, 169 Tubercled blossom, 130 Tubificidae, 20 Tufted hair, 119 Turbellaira, 20 Turk's cap lily, 120 Turkeybeard, eastern, 123 Turtle alligator snapping, 127 southern painted, 127 Turtlehead red, 118 rose, 118 Tvetenia bavarica gp., 22 Tvetinia discloripes gp., 22 Twayblade kidney-leaf, 120 Loesel's, 120 southern, 120 Twistedstalk, rosy, 122 Tympanuchus cupido, 130 Typha latifolia, 23-27 Tyto alba, 128, 163–164 Uenoidae, 21

Uenoidae, 21 Ulmus serotina, 123 Umbel-like sedge, 118 Umbra limi, 126 Umbrella-wort, pale, 120 Upland privet, 119 Upland sandpiper, 127 Urbanus proteus, 86

Ursus americanus, 128 Virginia-mallow, 122 Wedgescale, swamp, 122 Utricularia macrorhiza, 123 Vitamin E, 170–171 Wehrle's salamander, 127 effect on Cathepsin B, 170-171 WEIBEL, CHARLES, 165, 166 effect on Cathepsin L, 170-171 WESSEL, MARK V., 146 Vallisneria american, 123 effect on cysteine proteases, 170-West Virginia white, 87 VANARNUM, AARON, 165, 166, Western dwarf dandelion, 120 VITATOE, LEIGH ANNE, 167 Vanessa atalanta, 87 Western false gromwell, 121 Vitis labrusca, 123 Western mud snake, 127 V. cardui, 87 V. rupestris, 123 V. virginiensis, 87 Western pigmy rattlesnake, 127 Vitrinizonites latissimus, 124 Variable-leaved heartleaf, 119 Western ribbon snake, 127 Volatile emissions Western sand darter, 125 Varicose rocksnail, 123 biological effects of, 167-168 Variegated fritillary, 87 Westland size, effect on Canada from cut turf, 167-168 Goose, 46-49 Vascular plants; 117–123 Veery, 55 Wetlands, constructed, 23-29 Kentucky red-backed, 128 Wharton's dewberry, 122 Veneroida, 20 meadow, 164 White admiral, 87 Verbena, rose, 119 Vermivora bachmanii, 130 prairie, 163, 164 White catspaw, 130 woodland, 163, 164 V. chrysoptera, 50, 53, 128 White fringeless orchid, 121 Volvocales, 34 White heath aster, 117 V. pinus, 55 Vernonia noveboracensis, 123 White laldy's slipper, small, 118 Veronica americana, 123 Wabash riffleshell, 130 White peacock, 87 White rattlesnake-root, 121 Vertigo WALCK, JEFFREY L., 63 cupped, 124 Wallengrenia egeremet, 86 White snakeroot, 167 White snakeroot, Lucy Braun's, 117 delicate, 124 W. otho, 86 Vertigo bollesiana, 124 Walnut, white, 120 White walnut, 120 V. clappi, 124 Walter's violet, 123 White wartyback, 130 Vesper sparrow, 128 Warbler White Vetchling peavine, 120 cabbage, 87 Bachman's, 130 Viburnum lentago, 30–33 black-and-white, 56 checkered, 87 V. molle, 123 black-throated blue, 54 West Virginia, 87 V. nudum, 123 black-throated blue, 55 White-cedar, northern, 123 V. prunifolium, 30-32 black-throated blue, 56 White-eyed vireo, 53 White-haired goldenrod, 122 V. rafinesquianum var. rafinesquian-Blackburnian, 50, 53, 55, 127 White-leaved mountain-mint, 121 um, 123 blue-winged, 55 V. rufidulum, 30-32 Canada, 50, 53, 55, 128 White-m, 87 Whitelip, big-tooth, 123 Viburnum, possum haw, 123 cerulean, 50, 53 Viceroy, 87 chestnut-sided, 54-56 Whitelip, striped, 124 Villosa fabalis, 124 WHITEMAN, HOWARD, 171 golden-winged, 50, 53, 55, 128 V. lienosa, 124 hooded, 54-56 Whitewashed rabdotus, 123 V. ortmanni, 124 Whitlow- wedge-leaf, 119 yellow-rumped, 53 Whorled aster, 119 V. trabalis, 124 WARNER, RICHARD C., 23 Whorled horse-balm, 118 V. vanuxemensis, 124 Wartyback, white, 130 Viola septemloba var. egglestonii, Wastewater treatment, 23-29 Wild honeysuckle, 120 123 Water hickory, 118 Wild Indigo duskywing, 86 V. walteri, 123 Water locust, 119 Wild indigo Violet Water snake blue, 117 Eggleston's, 123 broad-banded, 127 cream, 117 Walter's, 123 yellow, 117 copperbelly, 127 Vireo bellii, 128 Wild lily-of-the-valley, 120 Mississippi green, 127 V. flavifrons, 53 Water stitchwort, 122 Wild rice V. griseus, 53 Water-milfoil Indian, 123 V. olivaceus, 53, 54 southern, 123 broadleaf, 121 Wild rye, Svenson's, 119 V. solitarius, 53, 54 cutleaf, 121 Willow, 55 Vireo Water-pennywort, American, 119 Bell's, 128 Water-plantain spearwort, 121 peachleaf, 122 blue-headed, 55 Water-purslane, 119 pussy, 122 Wilsonia canadensis, 50, 53, 128 red-eyed, 54, 55 Waterleaf, Virginia, 119 W. citrina, 53 white-eyed, 53 Waterweed, 119 Virginia big-eared bat, 128 Weak stellate sedge, 118 Winged mapleleaf, 130 Virginia bladetooth, 123 Weasel, least, 128 Winter wren, 55 Virginia bunchflower, 120 Webbhelix multilineata, 124 Wintergreen, American, 121 Virginia pine, 55 WEBSTER, CARL D., 166 Wire fern moss, 117 Virginia spiraea, 122 WECKMAN, TIMOTHY J., 30 Witch northern, 119 Virginia waterleaf, 119 Wedge-leaf whitlow, 119

gray, 130 red, 130 Wood fern southern shield, 119 spinulose, 119 Wood lily, 120 Wood sorrel, Price's yellow, 121 Wood thrush, 56 Wood-nymph, common, 87 Woodland beak-rush, 122 Woodland vole, 163, 164 Woodpecker ivory-billed, 130 red-cockaded, 128 Woodsia appalachiana, 123 Woodsia, mountain, 123 Woolly sedge, 118 Wormaldia prob. moesta, 21 Wren Bewick's, 128

Carolina, 56

sedge, 127

winter, 55 WRIGHT, DONALD J., 105 Wrinkled button, 123 Xanthidium antilopaeum, 40, 42 Xerophyllum asphodeloides, 123 Xyris difformis, 123

Yandell, David Wendel, M. D., 77–85
Yehl skipper, 86
Yellow blossom, 130
Yellow evening primrose, 118
Yellow gentian, 119
Yellow nodding ladies'-tresses, 122
Yellow screwstem, 117
Yellow water I., 27
Yellow wild indigo, 117
Yellow wood sorrel, Price's, 121
Yellow, little, 87
Yellow-bellied sapsucker, 53
Yellow-billed cuckoo, 53

Yellow-breasted chat, 53 Yellow-crested orchid, 121 Yellow-crowned night-heron, 127 Yellow-eye, Carolina, 123 Yellow-poplar, 54, 56 Yellow-rumped warbler, 53 Yellowthroat, common, 53 Yew, Canadian, 123 Yugus, 18 Yugus sp., 20

Zabulon skipper, 86 Zantedeschia aethiopica, 27 Zarucco duskywing, 86 Zebra swallowtail, 87 Zenaida macroura, 53 Zenaida macroura, 53 Zinc, effects of on rats, 108–114 Zizania palustris var. interior, 123 Zizaniopsis miliacea, 123 Zoology & Entomology, 171 Zygnema decussatum, 36, 38 Zygnematales, 36





# NEWS



The Morehead Electronic Journal of Applications in Mathematics (MEJAM) is a new interdisciplinary journal sponsored by Morehead State University, Morehead, Kentucky. The goal of MEJAM is to provide a refereed outlet for undergraduate students in any discipline to publish quality papers and see the results quickly. MEJAM accepts papers that are outside the realm of the typical undergraduate curriculum and that emphasize the applications of mathematics while maintaining significant mathematical interest. Papers may be historical, expository, or completely original in nature but must adhere to strict academic standards and must emphasize some aspect of the applications of mathematics. Papers from all disciplines will be considered for publication. More information about the journal and instructions for submissions can be found on the journal's website at http://www.morehead-st.edu/colleges/science/math/mejam/.

The Kentucky Academy of Science is seeking to complete its set of *Transactions of the Kentucky Academy of Science*. Various issues prior to 1985 are needed. Anyone willing to donate back issues or to sell them at a reasonable price should get in touch with the editor at thieretj@nku.edu.

## NOTE

Now available: **The Butterflies and Moths (Lepidoptera) of Kentucky: An Annotated Checklist.** 1999. Charles V. Covell Jr. Kentucky State Nature Preserves Commission Technical Series 6. 220 pp. Lists and gives localities, flight records, and remarks on the 2388 species of butterflies and moths known from Kentucky up to the time of publication. This work represents the author's study of butterflies and moths since his arrival at the University of Louisville in 1964; it includes records from collections made before and since that date. Order from Kentucky State Nature Preserves Commission, 801 Schenkel Lane, Frankfort, KY 40601-1403; \$17.00 postpaid. Make check payable to "Lepbook."

# **NEWS**

The 2001 annual meeting of the Kentucky Academy of Science will be held jointly with the Tennessee Academy of Science on Thursday—Saturday, 29–30 November and 1 December 2001, at Middle Tennessee State University (MTSU), Murfreesboro, Tennessee. There will be a reception and symposium at the Garden Plaza Hotel in Murfreesboro on Thursday evening. Friday technical sessions will be help on the MTSU campus in the Keathley University Center and James Union Building. Technical sessions will conclude on Saturday in the Keathley University Center.

## CONTENTS

## ARTICLES

Conservation Status and Nesting Biology of the Endangered Duskytail Darter, Etheostoma Percnurum, in the Big South Fork of the Cumberland River, Kentucky. David J. Eisenhour and Brooks M. Burr	67
Scientists of Kentucky David Wendel Yandell, M. D. Nancy Disher Baird	77
A Field Checklist of Kentucky Butterflies. Charles V. Covell Jr	86
Notes on North American Elymus Species (Poaceae) with Paired Spikelets: I. E. macgregorii sp. nov. and E. glaucus ssp. mackenzii comb. nov. Julian J. N. Campbell	88 *
Proterometra macrostoma (Digenea: Azygiidae): Distome Emergence From the Cercarial Tail and Subsequent Development in the Definitive Host. Ronald Rosen, Kelly Adams, Emilia Boiadgieva, and Jessica Schuster	99
New State Records and New Available Names for Species of Kentucky Moths (Insecta: Lepidoptera). Charles V. Covell Jr., Loran D. Gibson, and Donald J. Wright	105
Comparative Effects of Zinc, Lead, and Cadmium on Body and Tissue Weights of Weanling, Adult, and Aged Rats. F. N. Bebe and Myna Panemangalore	108
Rare and Extirpated Biota of Kentucky. Kentucky State Nature Preserves Commission	115
First Observations with the Morehead Radio Telescope, Morehead State University, Morehead, Kentucky. Benjamin K. Malphrus, Michael S. Combs, Michael A. James, D. Shannon Murphy, D. Kevin Brown, Jeff Kruth, and R. Douglas Kelly.	133
Agrimonia (Rosaceae) in Kentucky with Notes on the Genus. Mark V. Wessel and John W. Thieret	146
NOTE	
Barn Owl (Tyto alba) Feeding Habits at Yellowbank Wildlife Management Area, Breckinridge County, Kentucky. Anita T. Morzillo, Hetti A. Brown, Wayne E. Thogmartin, and Jennifer H. Herner-Thogmartin	163
Abstracts of Some Papers Presented at the 1999 Meeting of the Kentucky Academy of Science	165
List of Reviewers for Volume 61	173
Index to Volume 61	174